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<b>(21) International Application Number:</b> PCT/EP94/01671 <b>(22) International Filing Date:</b> 20 May 1994 (20.05.94)  <b>(30) Priority Data:</b> P 43 17 596.1      24 May 1993 (24.05.93)      DE  <b>(71) Applicant (for all designated States except US):</b> HOECHST SCHERING AGREVO GMBH [DE/DE]; Gerichtstrasse 27, D-13347 Berlin (DE).  <b>(72) Inventors; and</b> <b>(75) Inventors/Applicants (for US only):</b> HESSE, Holger [DE/DE]; Brundhildstrasse 9, D-10829 Berlin (DE). MÜLLER- RÖBER, Bernd [DE/DE]; Siemensstrasse 71, D-12247 Berlin (DE).		<b>(81) Designated States:</b> CA, HU, JP, RU, UA, US, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).  <b>Published</b> <i>Without international search report and to be republished upon receipt of that report.</i>
<b>(54) Title:</b> DNA SEQUENCES AND PLASMIDS FOR THE PREPARATION OF SUGAR BEET WITH CHANGED SUCROSE CONCENTRATION  <b>(57) Abstract</b>  DNA sequences and plasmids are described, that by integration in a plant genome of sugar beet change the sucrose concentration, as well as transgenic plants that by introduction of the DNA sequences of the invention causes changes in sugar concentration.		

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Title: DNA sequences and plasmids for the preparation of sugar beet with changed sucrose concentration

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Field of the invention

The present invention relates to DNA sequences and plasmids, containing these DNA sequences, which by integration into the genome of a sugar-beet plant, changes  
10 the sugar metabolism of the plant to be changed. The invention also relates to transgenic plants formed with the help of these sequences.

Sucrose is of central importance for the plant and serves  
15 many functions. For the long distance transport of photoassimilates and/or energy between various organs in plants, sucrose is almost exclusively used. The sucrose, which is transported in a specific heterotrophic organ, determines the growth and the development of this organ.  
20 Thus it is known, e.g. from EP 442 592, that transgenic plants, in which the transport away of the sucrose from the exporting leaves is inhibited by expression of an apoplastic invertase, shows a strong reduction in the growth of e.g. roots or tubers in the case of potato  
25 plants. For tobacco plants, the principal importance of sucrose as the central function for the long distance transport of energy carriers within the plant is described (von Schaewen et al, 1990, EMBO J 9: 3033-3044).

30 Further it is also known from EP 455 316 that DNA sequences present on plasmids, after introduction in a plant genome of a potato plant can affect the starch biosynthesis as well altering the amount and composition of the protein in the potato tubers.

35

Whilst it has known that a reduction of the amount of sucrose imported in the heterotrophic organs, such as tubers and seeds, leads to loss of yield, it is not known whether an increase in the amount of sucrose in the photosynthetically active parts of the plant, mainly the leaves, leads to a better supply of heterotrophic organs and thus to an increase in yield.

Besides sucrose and/or the hexoses, glucose and fructose, derived from sucrose, have the property of protection of plants against frost damage at low temperatures. Frost damage is one of the main limiting factors in agricultural productivity in the northern hemisphere. Temperatures below freezing lead to the formation of ice crystals. Since the growing ice crystals consist of pure water, water is abstracted from the cells as the temperature falls.

This dehydration has at least two potential damaging results:

1. All dissolved substances within a cell are strongly concentrated and the cell contracts following the loss of water. Highly concentrated salts and organic acids lead to membrane damage.

2. With rehydration from dew, the previously contacted cells reexpand. The cell membrane also expands again. The volume expansion puts a heavy mechanical load on the membrane.

It is thus clear that a freezing/dew cycle can lead to severe membrane damage of the cells and thus to damage to the plant.

It thus appears worth trying to hinder the freezing. One

possible strategy is the increased formation of osmotically active substances in the cytosol of plant cells. This should lead to a lowering of the freezing point. Osmotically active substances include sucrose  
5 and/or the two hexoses derived from sucrose.

The increased formation of sucrose and/or the two hexoses at low temperatures is desirable in the growing plant. Another situation can exist in the harvested parts of a  
10 plant, especially in storage.

In relation to the economic aspects, sucrose thus possesses two especially important functions:

- 1 as the transport form for the distant transport of  
15 photoassimilates,
- 2 as an osmotically active substance with the desirable activity of lowering the freezing point in intact, growing plants.

20 The biosynthesis pathways for the formation of sucrose, either from the primary photosynthesis products (in the leaf) or by breakdown of starch (in the storage organs e.g. of potatoes), are known.

25 It is however, not known how and in what way changes of the carbohydrate concentration in sugar beet can be achieved since it is not possible to use even very similar genes such as for example genes that code for a sucrose synthase, ADP-glucose pyrophosphorylase or sucrose  
30 phosphate synthase of the potato with satisfactory success for the preparation of sugar beet with changed sucrose concentration. An exact analysis and determination of the DNA sequences or sequence fragments for the sugar beet is thus required.

To change the sugar concentration in sugar beet, DNA sequences are now provided which code for the small and large subunit of the ADP glucose pyrophosphorylase, the sucrose synthase and the sucrose phosphate synthase of sugar beet (Seq. ID No 1-4).

These DNA sequences can be introduced into plasmids and in this way combined with steering elements for expression in eukaryotic cells. Such steering elements are on the one hand transcription promoters and on the other hand transcription terminators. Each plasmid comprises:

- a) a suitable promoter which ensures that the coding sequence meets a suitable time point or in the specified developments in the transgenic plant or in determined genes of transgenic plants;
- b) at least a coding sequence for sugar beet that
  - i) is so coupled to the promoter that the formation of an RNA is allowed which is into a protein, whereby the protein demonstrates an enzymatic activity which leads to a change of the sucrose concentration in the plant, or
  - ii) which is so coupled to the promoter that the coding part is read, which leads to the formation of a so-called anti-sense RNA which under-expresses the formation of the protein coded from an endogenous gene in the plant, that is involved in the sucrose biosynthesis; and
- c) A non-coding termination sequence that contains the signal for the termination and polyadenylation of the transcript.

## 5

The coding sequences named under b) are the sequences that code for the large and small subunit of the ADP glucose pyrophosphorylase, for the sucrose phosphate synthase and for the sucrose synthase of sugar beet.

## 5

The large subunit of the ADP-glucose-pyrophosphorylase has the following nucleotide sequence (Seq. ID No. 1):

CAAAAGAAAA ACTTCCCATT TCTACTTCTT TGCACAATAT AATTTCCCAC	0050
CAATTTTCTTCT TTAAATTTCT CACTTTCATT TAATCAGTTT TCAGCAACAT	0100
TCTGATACTC GACAACCCAC TTTCTGTTCT CCCAAGATTC CAAACCTCTG	0150
ATTCTCATTC CACTAATATT TTTGCTTATT TTTTTTCTGG ATTTAAAGAA	0200
AAGCT ATG GAT GCA AGT GCA GCA GCC ATA AAT GTC AAT GCC CAT	0243
Met Asp Ala Ser Ala Ala Ala Ile Asn Val Asn Ala His	
5 10	
TTA ACA GAA GTT GGA AAG AAA CGT TTT TTA GGA GAG AGA ATC AGT	0288
Leu Thr Glu Val Gly Lys Lys Arg Phe Leu Gly Glu Arg Ile Ser	
15 20 25	
CAA AGT TTG AAG GGT AAA GAT CTG AGA GCT CTG TTT TCA AGA ACT	0333
Gln Ser Leu Lys Gly Lys Asp Leu Arg Ala Leu Phe Ser Arg Thr	
30 35 40	
GAG AGC AAG GGT AGA AAT GTC AAT AAA CCT GGG GTT GCA TTT TCT	0378
Glu Ser Lys Gly Arg Asn Val Asn Lys Pro Gly Val Ala Phe Ser	
45 50 55	
GTT CTC ACC TCA GAT TTT AAT CAA AGT GTT AAA GAA TCT TTG AAA	0423
Val Leu Thr Ser Asp Phe Asn Gln Ser Val Lys Glu Ser Leu Lys	
60 65 70	

6

TAT GAG CCA GCA TTA TTT GAA TCT CCA AAA GCT GAC CCA AAA AAT 0468  
Tyr Glu Pro Ala Leu Phe Glu Ser Pro Lys Ala Asp Pro Lys Asn

75

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85

GTG GCT GCA ATT GTG CTG GGT GGT GGT GCT GGG ACT CGC CTC TTT 0513  
Val Ala Ala Ile Val Leu Gly Gly Gly Ala Gly Thr Arg Leu Phe

90

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100

CCT CTT ACT AGC AGG AGA GCT AAG CCA GCA GTG CCA ATT GGA GGG 0558  
Pro Leu Thr Ser Arg Arg Ala Lys Pro Ala Val Pro Ile Gly Gly

105

110

115

TGT TAC AGG CTG ATT GAT GTG CCT ATG AGC AAC TGC ATC AAC AGT 0603  
Cys Tyr Arg Leu Ile Asp Val Pro Met Ser Asn Cys Ile Asn Ser

120

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GGC ATT AGA AAG ATT TTC ATT CTT ACC CAG TTC AAT TCG TTT TCG 0648  
Gly Ile Arg Lys Ile Phe Ile Leu Thr Gln Phe Asn Ser Phe Ser

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CTT AAT CGT CAT CTT GCT CGA ACC TAT AAT TTT GGA GAT GGT GTG 0693  
Leu Asn Arg His Leu Ala Arg Thr Tyr Asn Phe Gly Asp Gly Val

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AAT TTT GGG GAT GGC TTT GTG GAG GTT TTT GCT GCT ACA CAA ACA 0738  
Asn Phe Gly Asp Gly Phe Val Glu Val Phe Ala Ala Thr Gln Thr

165

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CCT GGA GAA TCA GGA AAG AAA TGG TTC CAG GGC ACC GCT GAT GCA 0783  
Pro Gly Glu Ser Gly Lys Lys Trp Phe Gln Gly Thr Ala Asp Ala  
180 185 190

GTA AGA CAG TTT TTC TGG GCA TTT GAG GAT TCC AAA TCC AAG GAT 0828  
Val Arg Gln Phe Phe Trp Ala Phe Glu Asp Ser Lys Ser Lys Asp  
195 200 205

GTC GAG CAT ATA GTT ATT TTA TCC GGT GAT CAT CTT TAC CGA ATG 0873  
Val Glu His Ile Val Ile Leu Ser Gly Asp His Leu Tyr Arg Met  
210 215 220

GAT TAC ATG AGT TTT TGG CAG AAG CAC ATT GAC ACC AAT GCT GAT 0918  
Asp Tyr Met Ser Phe Trp Gln Lys His Ile Asp Thr Asn Ala Asp  
225 230 235

ATT ACA GTG TCA TGC ATA CCC ATG GAT GAC AGC CGT GCA TCG GAT 0963  
Ile Thr Val Ser Cys Ile Pro Met Asp Asp Ser Arg Ala Ser Asp  
240 245 250

TAT GGG CTG ATG AAG ATT GAT CAC ACT GGA CGC ATT GTC CAT TTT 1008  
Tyr Gly Leu Met Lys Ile Asp His Thr Gly Arg Ile Val His Phe  
255 260 265

GCA GAA AAA CCC AAG GGT TCT GAT CTA ACA GCA ATG CAA GTA GAT 1053  
Ala Glu Lys Pro Lys Gly Ser Asp Leu Thr Ala Met Gln Val Asp  
270 275 280

ACA ACT GTT CTT GGG CTC TCT GAC CTT GAA GCT ATG TCA AAT CCA 1098  
Thr Thr Val Leu Gly Leu Ser Asp Leu Glu Ala Met Ser Asn Pro  
285 290 295

TAT ATT GCA TCA ATG GGT GTT TAT GTC TTT CGA ACG GAT GTT CTT 1143  
Tyr Ile Ala Ser Met Gly Val Tyr Val Phe Arg Thr Asp Val Leu  
300 305 310

ATG GAG CTT CTC AAT CGA AAA TAC CCT TCA AGC AAT GAT TTT GGC 1188  
Met Glu Leu Leu Asn Arg Lys Tyr Pro Ser Ser Asn Asp Phe Gly  
315 320 325

TCT GAG ATT ATT CCT TCA GCT GTA GGA GAG TCT AAT GTT CAG GCA 1233  
Ser Glu Ile Ile Pro Ser Ala Val Gly Glu Ser Asn Val Gln Ala  
330 335 340

TAT CTA TTT AAT GAC TAC TGG GAG GAT ATC GGA ACC ATA AAG TCT 1276  
Tyr Leu Phe Asn Asp Tyr Trp Glu Asp Ile Gly Thr Ile Lys Ser  
345 350 355

TTC TTT GAT TCC AAT TTG GCC CTT ACA CAA CAG CCT CCC AAG TTT 1323  
Phe Phe Asp Ser Asn Leu Ala Leu Thr Gln Gln Pro Pro Lys Phe  
360 365 370

GAA TTC TAC GAT CCA AAA ACA CCT TTT TAT ACA TCT GCA AGA TTT 1368  
Glu Phe Tyr Asp Pro Lys Thr Pro Phe Tyr Thr Ser Ala Arg Phe  
375 380 385

CTG CCT CCT ACA AAA GTC GAC AGG TGC AAG ATT GTC GAT TCC ATT 1413  
Leu Pro Pro Thr Lys Val Asp Arg Cys Lys Ile Val Asp Ser Ile  
390 395 400

GTA TCC CAT GGT TGT TTT CTA CAG GAG TCT AGC ATC CAA CAT TCC 1458  
Val Ser His Gly Cys Phe Leu Gln Glu Ser Ser Ile Gln His Ser  
405 410 415

ATT GTT GGT GTT CGC TCA AGA TTA GAG TCC GGG GTT GAG TTC CAG 1503  
Ile Val Gly Val Arg Ser Arg Leu Glu Ser Gly Val Glu Phe Gln  
420 425 430

GAC ACC ATG ATG ATG GGC GCA GAT TAC TAT CAA ACT GAA TCA GAA 1548  
Asp Thr Met Met Met Gly Ala Asp Tyr Tyr Gln Thr Glu Ser Glu  
435 440 445

ATT GCT TCT CTG CTT GCT GAG GGA AAG GTT CCT GTT GGT GTC GGA 1593  
Ile Ala Ser Leu Leu Ala Glu Gly Lys Val Pro Val Gly Val Gly  
450 455 460

CAG AAT ACC AAA ATA AAG AAT TGC ATA ATT GAC AAG AAC GCC AAA 1638  
Gln Asn Thr Lys Ile Lys Asn Cys Ile Ile Asp Lys Asn Ala Lys  
465 470 475

ATT GGA AAA GAT GTG GTA ATC GCA AAC ACG GAT GGT GTT GAG GAA 1683  
Ile Gly Lys Asp Val Val Ile Ala Asn Thr Asp Gly Val Glu Glu  
480 485 490

10

GCA GAT AGA CCA AAT GAA GGC TTT TAC ATC AGG TCG GGC ATT ACC 1728  
 Ala Asp Arg Pro Asn Glu Gly Phe Tyr Ile Arg Ser Gly Ile Thr  
 495 500 505

ATC ATT TTG AAG AAC GCA ACC ATA CAA GAC GGT CTT GTG ATT TAG 1773  
 Ile Ile Leu Lys Asn Ala Thr Ile Gln Asp Gly Leu Val Ile End  
 510 515 520

ATTTAATCAT AACCTCATTA GAAAGAAATA ATTTTGCATG ATTTTCCTTTT 1823

CATGTAACCT AAACCTGGCTA AACCACGAGG TTTTCTCATC TGTATATATA 1873

ATATGTCTAT AACTATGGAT AATCTTAATA AAAAAAAAAA AAAAAAAAAA 1923

A 1924

The small subunit of the ADP-glucose-pyrophosphatase has  
 the following nucleotide sequence (Seq. ID No. 2):

GG ATA ACT GTG CCA TCA ACC TCC TCA AAG AAC CTC CAA AAT AGC 0044  
 Ile Thr Val Pro Ser Thr Ser Ser Lys Asn Leu Gln Asn Ser  
 5 10

CTC GCA TTC TCC TCT TCT TCT CTC TCC GGC GAC AAA ATT CAA ACG 0089  
 Leu Ala Phe Ser Ser Ser Ser Leu Ser Gly Asp Lys Ile Gln Thr  
 15 20 25

ACG TCA TTT CTC AAC CGC CGA TAT TGT AGA ATC TCT TCT AGA GCT 0134  
 Thr Ser Phe Leu Asn Arg Arg Tyr Cys Arg Ile Ser Ser Arg Ala  
 30 35 40

11

CCG ATT GTT GTC TCT CCC AAA GCT GTT TCT GAT TCT AAG AAT TCG 0179  
Pro Ile Val Val Ser Pro Lys Ala Val Ser Asp Ser Lys Asn Ser  
45 50 55

CAG ACT TGT CTT GAC CCT GAA GCC AGC CGT AGT GTT CTT GGT ATT 0224  
Gln Thr Cys Leu Asp Pro Glu Ala Ser Arg Ser Val Leu Gly Ile  
60 65 70

ATA CTT GGA GGT GGT GCT GGT ACA CGT CTT TAC CCG TTG ACT AAG 0269  
Ile Leu Gly Gly Gly Ala Gly Thr Arg Leu Tyr Pro Leu Thr Lys  
75 80 85

AAG AGA GCC AAA CCA GCC GTG CCA CTC GGT GCT AAT TAT AGG CTT 0314  
Lys Arg Ala Lys Pro Ala Val Pro Leu Gly Ala Asn Tyr Arg Leu  
90 95 100

ATT GAT ATC CCA GTG AGC AAT TGT TTG AAC AGT AAT ATT TCC AAA 0359  
Ile Asp Ile Pro Val Ser Asn Cys Leu Asn Ser Asn Ile Ser Lys  
105 110 110

ATA TAT GTT CTT ACA CAA TTC AAT TCT GCT TCT CTG AAT CGT CAT 0404  
Ile Tyr Val Leu Thr Gln Phe Asn Ser Ala Ser Leu Asn Arg His  
115 120 125

CTT TCG CGG GCA TAT GCT AGC AAC ATG GGA GGA TAC AAA AAT GAG 0449  
Leu Ser Arg Ala Tyr Ala Ser Asn Met Gly Gly Tyr Lys Asn Glu  
130 135 140

GGG TTT GTA GAA GTT CTT GCT GCT CAG CAA AGT CCA GAG AAT CCA 0494  
Gly Phe Val Glu Val Leu Ala Ala Gln Gln Ser Pro Glu Asn Pro  
145 150 155

12

AAC TGG TTT CAG GGT ACA GCT GAT GCT GTT AGG CAA TAT CTG TGG 0539  
Asn Trp Phe Gln Gly Thr Ala Asp Ala Val Arg Gln Tyr Leu Trp  
160 165 170

CTT TTC GAA GAG CAC AAT GTT CTT GAG TAC TTG ATT CTT GCT GGT 0584  
Leu Phe Glu Glu His Asn Val Leu Glu Tyr Leu Ile Leu Ala Gly  
175 180 185

GAC CAT TTG TAT CGA ATG GAT TAT GAA AGA TTT GTC CAA GCT CAC 0629  
Asp His Leu Tyr Arg Met Asp Tyr Glu Arg Phe Val Gln Ala His  
190 195 200

AGA GAA ACT GAT GCA GAC ATT ACT GTT GCT GCA TTG CCA ATG GAT 0674  
Arg Glu Thr Asp Ala Asp Ile Thr Val Ala Ala Leu Pro Met Asp  
205 210 215

GAA AAG CGT GCT ACT GCA TTT GGT TTG ATG AAA ATT GAT GAA GAA 0719  
Glu Lys Arg Ala Thr Ala Phe Gly Leu Met Lys Ile Asp Glu Glu  
220 225 230

GGA AGA ATT ATT GAG TTT GCC GAG AAA CCG AAA GGA GAA CAA TTG 0764  
Gly Arg Ile Ile Glu Phe Ala Glu Lys Pro Lys Gly Glu Gln Leu  
235 240 245

AAA GCT ATG AAG GTT GAT ACC ACA ATC CTG GGT CTG GAC GAT GAG 0809  
Lys Ala Met Lys Val Asp Thr Thr Ile Leu Gly Leu Asp Asp Glu  
250 255 260

13

AGA GCA AAA GAA ATG CCA TTC ATA GCC AGC ATG GGC ATA TAT GTT 0854  
 Arg Ala Lys Glu Met Pro Phe Ile Ala Ser Met Gly Ile Tyr Val  
 265 270 275

ATT AGC AAA GAT GTA ATG CTT AAT CTG CTT CGG GAG CAA TTT CCT 0899  
 Ile Ser Lys Asp Val Met Leu Asn Leu Leu Arg Glu Gln Phe Pro  
 280 285 290

GGT GCT AAT GAT TTT GGA AGT GAA GTT ATT CCA GGC GCC ACT TCC 0944  
 Gly Ala Asn Asp Phe Gly Ser Glu Val Ile Pro Gly Ala Thr Ser  
 295 300 305

ATA GGG TTG AGA GTC CAA GCT TAT TTG TAT GAT GGT TAC TGG GAG 0989  
 Ile Gly Leu Arg Val Gln Ala Tyr Leu Tyr Asp Gly Tyr Trp Glu  
 310 315 320

GAT ATT GGT ACC ATT GAA GCT TTT TAC AAT GCT AAC TTG GGA ATC 1034  
 Asp Ile Gly Thr Ile Glu Ala Phe Tyr Asn Ala Asn Leu Gly Ile  
 325 330 335

ACC AAA AAG CCG GTG CCA GAT TTT AGC TTC TAT GAT CGT TCA TCT 1079  
 Thr Lys Lys Pro Val Pro Asp Phe Ser Phe Tyr Asp Arg Ser Ser  
 340 345 350

CCA ATT TAT ACA CAA CCT CGG TAT TTG CCT CCT TCA AAG ATG CTT 1124  
 Pro Ile Tyr Thr Gln Pro Arg Tyr Leu Pro Pro Ser Lys Met Leu  
 355 360 365

14

GAT GCT GAT ATA ACT GAC AGC GTC ATC GGT GAA GGC TGT GTT ATT 1169  
Asp Ala Asp Ile Thr Asp Ser Val Ile Gly Glu Gly Cys Val Ile  
370 375 380

AAG AAC TGT AAG ATT CAT CAT TCT GTT ATC GGA CTT CGA TCT TGT 1214  
Lys Asn Cys Lys Ile His His Ser Val Ile Gly Leu Arg Ser Cys  
385 390 395

ATC TCG GAG GGT GCA ATC ATT GAG GAC ACA CTG TTG ATG GGA GCT 1259  
Ile Ser Glu Gly Ala Ile Ile Glu Asp Thr Leu Leu Met Gly Ala  
400 405 410

GAT TAT TAT GAG ACT GAT GCT GAT CGG AAA TTC CTG GCT GCT AAG 1304  
Asp Tyr Tyr Glu Thr Asp Ala Asp Arg Lys Phe Leu Ala Ala Lys  
415 420 425

GGT AGT GTA CCT ATT GGA ATT GGG AAT GCA CGT ATT GGG GAT GAT 1349  
Gly Ser Val Pro Ile Gly Ile Gly Asn Ala Arg Ile Gly Asp Asp  
430 435 440

GTC AAG ATT ATC AAC AGT GAC AAT GTA CAA GAA GCA GCA AGA GAA 1394  
Val Lys Ile Ile Asn Ser Asp Asn Val Gln Glu Ala Ala Arg Glu  
445 450 455

ACA GAC GGA TAC TTC ATA AAG AGC GGA ATA GTC ACT ATA ATC AAG 1439  
Thr Asp Gly Tyr Phe Ile Lys Ser Gly Ile Val Thr Ile Ile Lys  
460 465 470



15

GAC GCC ATG ATT CCA AGT GGA ACT GTA ATC TAG AAATGGAGCA 1482  
 Asp Ala Met Ile Pro Ser Gly Thr Val Ile End  
 475 480 485

TATAATAAAT ATCACTGCCT ATTTACAGTA CCTATCTGAG TCTCCCACCA 1532

TGACCCTTTG ATTCAATCTT TTAGTTATGT AAATATTTTT GGCTTTTGCG 1582

ATTTTGCCAT AAATTTGAAG AAGCGAGGAT TCAGGGACGA TAGTGCTATG 1632

AATTGGAAGA AAGGATTTGG GGGATATCTT TGTAAGACA TTTTGACTAC 1682

TGGGCACTAA AAATTTGGTA ATGCTATACC AAAATATATA AAAAGATCTT 1732

GCTGGGTTTT GTAAAAAAA AAAAAAAAAA A 1763

The sucrose phosphate - synthase has the following  
 nucleotide sequence (Seq. ID No. 3):

GGGCTGCAGG GAAGCTCTGA ACTTCAAAA ATG GCG GGA AAT GAT 0044  
 Met Ala Gly Asn Asp  
 5

TGG ATA AAC AGT TAT TTA GAG GCA ATT CTG GAT GTG GGT CCA GGA 0089  
 Trp Ile Asn Ser Tyr Leu Glu Ala Ile Leu Asp Val Gly Pro Gly  
 10 15 20

CTT GAT GAT GCA AAA TCA TCT TTG CTT TTG AGA GAA AGA GGC AGG 0134  
 Leu Asp Asp Ala Lys Ser Ser Leu Leu Leu Arg Glu Arg Gly Arg  
 25 30 35

TTT AGT CCT ACT CGT TAC TTT GTT GAA GAA GTT ATC ACT GGT TTT 0179  
 Phe Ser Pro Thr Arg Tyr Phe Val Glu Glu Val Ile Thr Gly Phe  
 40 45 50

16

GAT GAA ACC GAC CTT CAT CGT TCA TGG GTT CGG GCA CAA GCA ACA 0224  
 Asp Glu Thr Asp Leu His Arg Ser Trp Val Arg Ala Gln Ala Thr  
 55 60 65

AGG AGT CCT CAA GAG AGG AAT ACT AGA TTG GAG AAC ATG TGT TGG 0269  
 Arg Ser Pro Gln Glu Arg Asn Thr Arg Leu Glu Asn Met Cys Trp  
 70 75 80

AGA ATT TGG AAT TTG GCT CGT CAG AAG AAG CAG CTT GAG AAT GAA 0314  
 Arg Ile Trp Asn Leu Ala Arg Gln Lys Lys Gln Leu Glu Asn Glu  
 85 90 95

GAA GCT CAG CGG AAG ACA AAA CGT CGT ATG GAG CTT GAG AGG GGT 0359  
 Glu Ala Gln Arg Lys Thr Lys Arg Arg Met Glu Leu Glu Arg Gly  
 100 105 110

CGT CGA GAA GCA ACT GCT GAT ATG TCG GAG GAC TTA TCA GAA GGC 0404  
 Arg Arg Glu Ala Thr Ala Asp Met Ser Glu Asp Leu Ser Glu Gly  
 115 120 125

GAA AAG GAC ATT TCA GCT CAT GGT GAT AGC ACC CGT CCT AGA TTG 0449  
 Glu Lys Asp Ile Ser Ala His Gly Asp Ser Thr Arg Pro Arg Leu  
 130 135 140

CCA AGA ATA AAT TCT CTT GAT GCT ATG GAG ACA TGG ATT AGT CAA 0494  
 Pro Arg Ile Asn Ser Leu Asp Ala Met Glu Thr Trp Ile Ser Gln  
 145 150 155

CAA AAG GAA AAA AAA CTC TAC CTT GTT TTG ATA AGT CTT CAT GGT 0539  
 Gln Lys Glu Lys Lys Leu Tyr Leu Val Leu Ile Ser Leu His Gly  
 160 165 170

17

TTG ATA CGA GGT GAA AAC ATG GAA CTT GGC CGT GAT TCT GAT ACT 0584  
Leu Ile Arg Gly Glu Asn Met Glu Leu Gly Arg Asp Ser Asp Thr

175

180

185

GGT GGT CAG GTT AAG TAT GTG GTT GAG CTT GCA AGG GCT CTA GGT 0629  
Gly Gly Gln Val Lys Tyr Val Val Glu Leu Ala Arg Ala Leu Gly

190

195

200

TCG ATG CCA GGT GTT TAT AGA GTT GAT TTG CTA ACT AGG CAA GTT 0674  
Ser Met Pro Gly Val Tyr Arg Val Asp Leu Leu Thr Arg Gln Val

205

210

215

TCA TCT CCT GAC GTG GAT TGG AGT TAT GGG GAG CCT ACT GAG ATG 0719  
Ser Ser Pro Asp Val Asp Trp Ser Tyr Gly Glu Pro Thr Glu Met

220

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CTG AAT CCA AGG GAT TCC AAT GGT TTT GAT GAT GAT GAT GAT GAA 0764  
Leu Asn Pro Arg Asp Ser Asn Gly Phe Asp Asp Asp Asp Asp Glu

235

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245

ATG GGA GAG AGT AGT GGT GCT TAC ATT GTT CGT ATA CCA TTT GGG 0809  
Met Gly Glu Ser Ser Gly Ala Tyr Ile Val Arg Ile Pro Phe Gly

250

255

260

CCG AGG GAT AAG TAT ATC GCA AAA GAA GAG CTT TGG CCC TAT ATT 0854  
Pro Arg Asp Lys Tyr Ile Ala Lys Glu Glu Leu Trp Pro Tyr Ile

265

270

275

18

CCT GAA TTT GTT GAT GGT GCT CTA AAC CAC ATA GTT CAA ATG TCC 0899  
 Pro Glu Phe Val Asp Gly Ala Leu Asn His Ile Val Gln Met Ser  
 280 285 290

AAA GTT TTA GGT GAG CAA ATT GGT AGC GGG GAA ACA GTT TGG CCA 0944  
 Lys Val Leu Gly Glu Gln Ile Gly Ser Gly Glu Thr Val Trp Pro  
 295 300 305

GTT GCC ATT CAT GGA CAT TAT GCT GAT GCT GGT GAT TCT GCT GCT 0989  
 Val Ala Ile His Gly His Tyr Ala Asp Ala Gly Asp Ser Ala Ala  
 310 315 320

CTT CTT TCT GGT GGC CTA AAT GTT CCA ATG CTT TTA ACG GGG CAT 1034  
 Leu Leu Ser Gly Gly Leu Asn Val Pro Met Leu Leu Thr Gly His  
 325 330 335

TCT CTT GGC CGA GAC AAG TTA GAG CAG CTC CTC AAA CAG GGT CGA 1079  
 Ser Leu Gly Arg Asp Lys Leu Glu Gln Leu Leu Lys Gln Gly Arg  
 340 345 350

ATG TCT AAA GAT GAC ATA AAC AAT ACA TAC AAA ATA ATG CGT AGG 1124  
 Met Ser Lys Asp Asp Ile Asn Asn Thr Tyr Lys Ile Met Arg Arg  
 355 360 365

ATA GAA GCC GAA GAG TTA TCA CTT GAT GCC TCT GAG ATA GTC ATA 1169  
 Ile Glu Ala Glu Glu Leu Ser Leu Asp Ala Ser Glu Ile Val Ile  
 370 375 380

19

ACT AGT ACA AGA CAA GAA ATA GAA GAG CAA TGG CAC CTC TAT GAT 1214  
Thr Ser Thr Arg Gln Glu Ile Glu Glu Gln Trp His Leu Tyr Asp  
385 390 395

GGG TTT GAT CCT GTG CTA GAA CGT AAA CTC CGT GCT AGG ATG AAG 1259  
Gly Phe Asp Pro Val Leu Glu Arg Lys Leu Arg Ala Arg Met Lys  
400 405 410

CGT GGT GTA AGC TGT TAT GGA AGG TTC ATG CCC CGG ATG GTT GTT 1304  
Arg Gly Val Ser Cys Tyr Gly Arg Phe Met Pro Arg Met Val Val  
415 420 425

ATT CCT CCT GGA ATG GAA TTC AAT CAT ATT GTT CCA CAT GAG GGT 1349  
Ile Pro Pro Gly Met Glu Phe Asn His Ile Val Pro His Glu Gly  
430 435 440

GAT ATG GAT GGT GAA ACA GAA GAA ACT GAA GAG CAT CCT ACA TCA 1394  
Asp Met Asp Gly Glu Thr Glu Glu Thr Glu Glu His Pro Thr Ser  
445 450 455

CCT GAT CCA CCT ATC TGG GCT GAG ATT ATG CGC TTC TTT TCT AAA 1439  
Pro Asp Pro Pro Ile Trp Ala Glu Ile Met Arg Phe Phe Ser Lys  
460 465 470

CCA AGG AAG CCA ATG ATA CTT GCC CTT GCT AGG CCT GAC CCG AAG 1484  
Pro Arg Lys Pro Met Ile Leu Ala Leu Ala Arg Pro Asp Pro Lys  
475 480 485

20

AAG AAT ATC ACG ACT TTG GTC AAA GCA TTT GGA GAA TGC CGT CCA 1529  
Lys Asn Ile Thr Thr Leu Val Lys Ala Phe Gly Glu Cys Arg Pro  
490 495 500

CTA AGG GAG CTA GCT AAT CTT ACT CTT ATA ATG GGT AAC CGA GAT 1574  
Leu Arg Glu Leu Ala Asn Leu Thr Leu Ile Met Gly Asn Arg Asp  
505 510 515

GGT ATT GAC GAG ATG TCA AGC ACC AGT TCT TCA GTT CTC CTG TCA 1619  
Gly Ile Asp Glu Met Ser Ser Thr Ser Ser Ser Val Leu Leu Ser  
520 525 530

GTG CTT AAG CTA ATT GAT CAA TAC GAC CTT TAT GGT CAA GTA GCA 1664  
Val Leu Lys Leu Ile Asp Gln Tyr Asp Leu Tyr Gly Gln Val Ala  
535 540 545

TAC CCC AAA CAT CAC AAG CAA GCT GAT GTT CCT GAG ATT TAT CGT 1709  
Tyr Pro Lys His His Lys Gln Ala Asp Val Pro Glu Ile Tyr Arg  
550 555 600

TTG GCA GCA AAG ACA AAG GGA GTC TTT ATT AAT CCA GCT TTT ATT 1754  
Leu Ala Ala Lys Thr Lys Gly Val Phe Ile Asn Pro Ala Phe Ile  
605 610 615

GAG CCA TTT GGG CTG ACT CTA ATA GAG GCA GCA GCT CAT GGT TTA 1799  
Glu Pro Phe Gly Leu Thr Leu Ile Glu Ala Ala Ala His Gly Leu  
620 625 630

21

CCG ATG GTT GCT ACG AAA AAT GGA GGC CCT GTT GAT ATC CAG AGG 1844  
Pro Met Val Ala Thr Lys Asn Gly Gly Pro Val Asp Ile Gln Arg  
635 640 645

GTC CTT GAT AAT GGT CTT CTT GTG GAT CCT CAT GAG CAG CAG TCT 1889  
Val Leu Asp Asn Gly Leu Leu Val Asp Pro His Glu Gln Gln Ser  
650 655 660

ATT GCT ACT GCT TTG CTG AAG CTT GTT GCT GAT AAG CAA CTA TGG 1934  
Ile Ala Thr Ala Leu Leu Lys Leu Val Ala Asp Lys Gln Leu Trp  
665 670 675

ACA AAA TGC CAG CAA AAT GGA CTG AAA AAT ATT CAT CTC TAC TCT 1979  
Thr Lys Cys Gln Gln Asn Gly Leu Lys Asn Ile His Leu Tyr Ser  
680 685 690

TGG CCA GAG CAT TCG AAG ACA TAC CTA TCT CGA ATA GCC AGT TCG 2024  
Trp Pro Glu His Ser Lys Thr Tyr Leu Ser Arg Ile Ala Ser Ser  
695 700 705

AGA CAA AGG CAA CCA CAG TGG CAA AGA AGT AGT GAT GAA GGG CTT 2069  
Arg Gln Arg Gln Pro Gln Trp Gln Arg Ser Ser Asp Glu Gly Leu  
710 715 720

GAC AAT CAA GAG CCT GAA TCT CCA AGT GAT TCT TTA AGA GAT ATA 2114  
Asp Asn Gln Glu Pro Glu Ser Pro Ser Asp Ser Leu Arg Asp Ile  
725 730 735

22

AAG GAT ATA TCT CTA AAC CTT GAA GTT CTC GTT AGA CCG GAG AAA 2159  
Lys Asp Ile Ser Leu Asn Leu Glu Val Leu Val Arg Pro Glu Lys  
740 745 750

AGG GTG AAG ACG TTG AAA ATC TTG GGA TTG ATG ACA AAA GCA AAT 2204  
Arg Val Lys Thr Leu Lys Ile Leu Gly Leu Met Thr Lys Ala Asn  
755 760 765

TCG AGA ATG CTG TTA TGT TCA TGG TCT AAT GGT GTC CAT AAG ATG 2249  
Ser Arg Met Leu Leu Cys Ser Trp Ser Asn Gly Val His Lys Met  
770 775 780

CTT CGG AAG GCT CGG TTC TCT GAC AAA GTA GAT CAG GCT TCT AGT 2294  
Leu Arg Lys Ala Arg Phe Ser Asp Lys Val Asp Gln Ala Ser Ser  
785 790 795

AAA TAT CCA GCA TTT AGG AGG AGA AAA CTT ATA TAT GTT ATT GCT 2339  
Lys Tyr Pro Ala Phe Arg Arg Arg Lys Leu Ile Tyr Val Ile Ala  
800 805 810

GTA GAC GGG GAT TAT GAA GAT GGA CTT TTT GAT ATT GTT CCG AGG 2384  
Val Asp Gly Asp Tyr Glu Asp Gly Leu Phe Asp Ile Val Arg Arg  
815 820 825

ATA TTT GAT GCT GCT GGC AAG GAG AAG ATT GAA GGT TCC ATC GGG 2429  
Ile Phe Asp Ala Ala Gly Lys Glu Lys Ile Glu Gly Ser Ile Gly  
830 835 840



23

TTT ATA TTG TCA ACA TCC TAT TCT ATG CCC GAA ATT CAG AAC TAT 2474  
Phe Ile Leu Ser Thr Ser Tyr Ser Met Pro Glu Ile Gln Asn Tyr  
845 850 855

TTG CTA TCA AAA GGC TTC AAT CTT CAT GAT TTT GAT GCA TAT ATA 2519  
Leu Leu Ser Lys Gly Phe Asn Leu His Asp Phe Asp Ala Tyr Ile  
860 865 870

TGC AAC AGT GGG AGT GAG TTG TAC TAT TCA TCT TTG AAC TCA GAG 2564  
Cys Asn Ser Gly Ser Glu Leu Tyr Tyr Ser Ser Leu Asn Ser Glu  
875 880 885

GAG AGT AAT ATT ATA GCA GAT TCA GAT TAC CAT TCA CAC ATA GAG 2609  
Glu Ser Asn Ile Ile Ala Asp Ser Asp Tyr His Ser His Ile Glu  
890 895 900

TAC AGA TGG GGT GGA GAA GGC CTT AGA AGG ACT TTG CTT CGC TGG 2654  
Tyr Arg Trp Gly Gly Glu Gly Leu Arg Arg Thr Leu Leu Arg Trp  
905 910 915

GCA GCT TCC ATC ACA GAA AAA AAT GGT GAA AAC GAA GAA CAG GTT 2699  
Ala Ala Ser Ile Thr Glu Lys Asn Gly Glu Asn Glu Glu Gln Val  
920 925 930

ATT ACT GAA GAT GAA GAA GTT TCT ACG GGT TAT TGC TTT GCG TTT 2744  
Ile Thr Glu Asp Glu Glu Val Ser Thr Gly Tyr Cys Phe Ala Phe  
935 940 945

24

AAA ATA AAG AAC CAA AAT AAG GTT CCC CCT ACG AAG GAG CTC CGC 2789  
 Lys Ile Lys Asn Gln Asn Lys Val Pro Pro Thr Lys Glu Leu Arg  
 950 955 960

AAG TCA ATG AGG ATT CAA GCT CTT CGT TGC CAT GTG ATT TAC TGT 2834  
 Lys Ser Met Arg Ile Gln Ala Leu Arg Cys His Val Ile Tyr Cys  
 965 970 975

CAG AAC GGA TCT AAA ATG AAT GTG ATT CCA GTA CTA GCA TCC CGT 2879  
 Gln Asn Gly Ser Lys Met Asn Val Ile Pro Val Leu Ala Ser Arg  
 980 985 990

TCT CAA GCC CTC AGG TAT CTT TAT GTT CGT TGG GGA GTT GAG TTG 2924  
 Ser Gln Ala Leu Arg Tyr Leu Tyr Val Arg Trp Gly Val Glu Leu  
 995 1000 1005

TCG AAG ATG GTT GTC TTT GTT GGA GAA TGT GGT GAC ACA GAT TAT 2969  
 Ser Lys Met Val Val Phe Val Gly Glu Cys Gly Asp Thr Asp Tyr  
 1010 1015 1020

GAA GGC TTG CTT GGC GGG GTC CAT AAA ACC GTA ATA CTG AAG GGA 3014  
 Glu Gly Leu Leu Gly Gly Val His Lys Thr Val Ile Leu Lys Gly  
 1025 1030 1035

GTC TCC AAC ACT GCT TTA AGG TCT CTC CAT GCC AAC AGA AGT TAC 3059  
 Val Ser Asn Thr Ala Leu Arg Ser Leu His Ala Asn Arg Ser Tyr  
 1040 1045 1050

25

CCT CTT TCA CAT GTC GTG TCG CTT GAC AGC CCC AAT ATT GGC GAG 3104  
Pro Leu Ser His Val Val Ser Leu Asp Ser Pro Asn Ile Gly Glu  
1055 1060 1065

GTG AGC AAA GGG TGC AGC AGC TCC GAG ATC CAG TCC ATC GTC ACA 3149  
Val Ser Lys Gly Cys Ser Ser Ser Glu Ile Gln Ser Ile Val Thr  
1070 1075 1080

AAA CTC TCC AAA GCT TAA TCAGATATCT GCTGCTTTCT TTTGGGTAAG 3197  
Lys Leu Ser Lys Ala End  
1085

CAAGGTTTCA TCTTATATGA TTATATCATA AGATACTATA TAAGCACCTT 3247

ATTGGTAAGT CAGTCCCATA ATAATAATGT ACTTCAGAAC CACAATACTT 3297

AAAAGTTGGT TCAGTAGTGA TTAGTCTCAT AATAATCATA TAATTACACA 3347

TCCGCTGTTA ACTAGTGGTA ATATCTAAGC TCAACAATAA AGATGTAAAA 3397

TGCTAGTATG GAAATGAATT GCTAGCTGTT GATCTCTTTC CCTTTATTCT 3447

GTATTATTTC TTTCCTCATC TCATGTAAAA ACAATTTTCT GAAGGTGTAC 3497

AGTTTTTTCC CCTTATATAT CTGTATTATT TCTACTATTT TTTGTTTGTA 3547

AGAATATCCT CTCATCGAGG AGTGATAATT AAATAACCGG CTTGCTAAAT 3597

ATAAAGCTTA TTCGAGTTAA AAAAAAAAAA AAAAAAAAAA 3635

The sucrose-synthase has the following nucleotide sequence  
(Seq. ID No. 4):

CT GCA GGA GGG AAA CAA ATT CTT AGC GAT GGC CCG TTT AGC GAA	0044
Ala Gly Gly Lys Gln Ile Leu Ser Asp Gly Pro Phe Ser Glu	
5 10	
GTT CTT AGG TCT GCT CAG GAA GCA ATA GTT GTT CCT CCC TTT GTT	0089
Val Leu Arg Ser Ala Gln Glu Ala Ile Val Val Pro Pro Phe Val	
15 20 25	
GCT ATA GCA GTC CGT CCA AGA CCT GGA GTT TGG GAA TAT GTT CGT	0134
Ala Ile Ala Val Arg Pro Arg Pro Gly Val Trp Glu Tyr Val Arg	
30 35 40	
GTT AAT GTC TCT GAA CTG AAT GTG GAG CAG CTA ACT GTG TCT GAG	0179
Val Asn Val Ser Glu Leu Asn Val Glu Gln Leu Thr Val Ser Glu	
45 50 55	
TAT CTC CAT TTC AAG GAA GAA CTT GTG GAT GGA AAG GCT GAT GAC	0224
Tyr Leu His Phe Lys Glu Glu Leu Val Asp Gly Lys Ala Asp Asp	
60 65 70	
CAC TAT GTG CTT GAG CTT GAT TTC GAG CCT TTT AAT GAA TCA GTT	0269
His Tyr Val Leu Glu Leu Asp Phe Glu Pro Phe Asn Glu Ser Val	
75 80 85	
CCA CGT CCA ACT CGC TCT TCA TCA ATT GGT AAT GGT GTT CAG TTC	0314
Pro Arg Pro Thr Arg Ser Ser Ser Ile Gly Asn Gly Val Gln Phe	
90 95 100	
CTC AAT CGG CAC CTG TCA TCA AGC ATG TTC TGC AAC AAA GAT TGC	0359
Leu Asn Arg His Leu Ser Ser Ser Met Phe Cys Asn Lys Asp Cys	
105 110 115	

27

TTG GAG CCG TTA CTT GAT TTT CTT AGA GTG CAC AAA CAT AAA GGA 0404  
Leu Glu Pro Leu Leu Asp Phe Leu Arg Val His Lys His Lys Gly  
120 125 130

GTT GTC ATG ATG TTG AAT GAT CGG ATA CAG ACT ATC CAG CGT CTT 0449  
Val Val Met Met Leu Asn Asp Arg Ile Gln Thr Ile Gln Arg Leu  
135 140 145

CAG TCT GCA TTG TCT AAA GCT GAG GAT TAT CTT ATC AAA CTT CCA 0494  
Gln Ser Ala Leu Ser Lys Ala Glu Asp Tyr Leu Ile Lys Leu Pro  
150 155 160

GCA GAT ACA CCT TAC TCT GAG TTC GAA TTT GTA ATC CAA GGT ATG 0539  
Ala Asp Thr Pro Tyr Ser Glu Phe Glu Phe Val Ile Gln Gly Met  
165 170 175

GGT TTT GAA AGA GGC TGG GGT GAT ACT GCT GAA AGG GTT CTA GAA 0564  
Gly Phe Glu Arg Gly Trp Gly Asp Thr Ala Glu Arg Val Leu Glu  
180 185 190

ATG ATG CAT CTA CTA CTA GAT ATC CTT CAG GCT CCC GAT CCG TCT 0629  
Met Met His Leu Leu Leu Asp Ile Leu Gln Ala Pro Asp Pro Ser  
195 200 205

ACA TTA GAG ACA TTT CTG GGA AGA CTT CCC ATG GTG TTT AAT GTG 0674  
Thr Leu Glu Thr Phe Leu Gly Arg Leu Pro Met Val Phe Asn Val  
210 215 220

28

GTC ATT TTG TCT GTA CAT GGA TAT TTT GGA CAG GCA CAT GTG CTC 0719  
Val Ile Leu Ser Val His Gly Tyr Phe Gly Gln Ala His Val Leu  
225 230 235

GGC TTG CCT GAC ACT GGT GGG CAG ATA GTT TAT ATA CTT GAC CAA 0764  
Gly Leu Pro Asp Thr Gly Gly Gln Ile Val Tyr Ile Leu Asp Gln  
240 245 250

GTG CGG TCT CTG GAA CAT GAA ATG CTC CAA CGA ATA AAG AAG CAA 0809  
Val Arg Ser Leu Glu His Glu Met Leu Gln Arg Ile Lys Lys Gln  
255 260 265

GGA CTA GAT GTG ACT CCT AGA ATT CTT ATC GTG AGT CGG TTG ATT 0854  
Gly Leu Asp Val Thr Pro Arg Ile Leu Ile Val Ser Arg Leu Ile  
270 275 280

CCT GAC GCT AAA GGG ACC ACG TGC AAT CAA CGT ATG GAG AAA GTC 0899  
Pro Asp Ala Lys Gly Thr Thr Cys Asn Gln Arg Met Glu Lys Val  
285 290 295

AGT GGA ACA GAG CAT GCT AGT ATC CTG AGA GTT CCT TTC CGA TCA 0944  
Ser Gly Thr Glu His Ala Ser Ile Leu Arg Val Pro Phe Arg Ser  
300 305 310

GAG AAA GGA ATC CTC CGC AAA TGG ATA TCT AGA TTT GAT GTA TGG 0989  
Glu Lys Gly Ile Leu Arg Lys Trp Ile Ser Arg Phe Asp Val Trp  
315 320 325

29

CCT TAT TTA GAG ACC TTC ACT GAG GAT GCA GCT GGT GAA ATT ATT 1034  
Pro Tyr Leu Glu Thr Phe Thr Glu Asp Ala Ala Gly Glu Ile Ile  
330 335 340

GGC GAG TTG CAG GGT CGT CCA GAT CTG ATA ATT GGC AAC TAC AGC 1079  
Gly Glu Leu Gln Gly Arg Pro Asp Leu Ile Ile Gly Asn Tyr Ser  
345 350 355

GAT GGG AAT ATA GTT GCT TCT TTA TTG TCC CAC AAA ATG GGT GTC 1124  
Asp Gly Asn Ile Val Ala Ser Leu Leu Ser His Lys Met Gly Val  
360 365 370

ACC CAG TGC AAT ATA GCC CAT GCA TTG GAG AAA ACC AAG TAT CCA 1169  
Thr Gln Cys Asn Ile Ala His Ala Leu Glu Lys Thr Lys Tyr Pro  
375 380 385

GAT TCT GAT ATT TAC TGG AAA AGA TTT GAG GAC AAA TAT CAC TTC 1214  
Asp Ser Asp Ile Tyr Trp Lys Arg Phe Glu Asp Lys Tyr His Phe  
390 395 400

TCG TGT CAA TTT TCA GCT GAC TTG ATG GCA ATG AAT CAT GCT GAT 1259  
Ser Cys Gln Phe Ser Ala Asp Leu Met Ala Met Asn His Ala Asp  
405 410 415

TTC ATC ATT ACG AGT ACT TAC CAA GAG ATA GCT GGA ACG AAG AAT 1304  
Phe Ile Ile Thr Ser Thr Tyr Gln Glu Ile Ala Gly Thr Lys Asn  
420 425 430

30

ACT GTT GGT CAA TAT GAA AGC CAT AAG GCC TTT ACT TTT CCG GGG 1349  
Thr Val Gly Gln Tyr Glu Ser His Lys Ala Phe Thr Phe Pro Gly  
435 440 445

CTG TAT CGG GTG GTT CAC GGG ATT GAT GTC TTT GAT CCC AAG TTT 1394  
Leu Tyr Arg Val Val His Gly Ile Asp Val Phe Asp Pro Lys Phe  
450 455 460

AAT ATT GTC TCG CCA GGG GCA GAC ATG GCC ATC TAC TTC CCA TTT 1439  
Asn Ile Val Ser Pro Gly Ala Asp Met Ala Ile Tyr Phe Pro Phe  
465 470 475

TCA GAG AAG GAT GTC ACC TGT CTC ACT TCA CTT CAT AGA CTT ATA 1484  
Ser Glu Lys Asp Val Thr Cys Leu Thr Ser Leu His Arg Leu Ile  
480 485 490

GAG CAG CTC CTA TTC AAA CCT GAG CAG AAC GAA GAA CAC ATT GGT 1529  
Glu Gln Leu Leu Phe Lys Pro Glu Gln Asn Glu Glu His Ile Gly  
495 500 505

GTA TTA GAT GAT ACC TCA AAG CCA ATT ATA TTT TCC ATG GCG AGG 1574  
Val Leu Asp Asp Thr Ser Lys Pro Ile Ile Phe Ser Met Ala Arg  
510 515 520

CTA GAC CGT GTG AAG AAT ATA ACA GGG CTG GTA GAG TGC TAT GGC 1619  
Leu Asp Arg Val Lys Asn Ile Thr Gly Leu Val Glu Cys Tyr Gly  
525 530 535



31

AAG AAT GCG AAA CTC AGG GAA CTG GCA AAC CTG GTT GTA GTG GCT 1664  
Lys Asn Ala Lys Leu Arg Glu Leu Ala Asn Leu Val Val Val Ala  
540 545 550

GGG TAC AAT GAT GTA AAA AAG TCG AAT GAC AGG GAG GAA ATT GCC 1709  
Gly Tyr Asn Asp Val Lys Lys Ser Asn Asp Arg Glu Glu Ile Ala  
555 560 565

GAA ATC GAG AAG ATG CAC AGG CTT ATA CAG GAG TAT AAT TTA AGA 1754  
Glu Ile Glu Lys Met His Arg Leu Ile Gln Glu Tyr Asn Leu Arg  
570 575 580

GGA CAA TTT CGC TGG ATT GCT TCT CAA ACA AAT AGA GTA CGA AAT 1799  
Gly Gln Phe Arg Trp Ile Ala Ser Gln Thr Asn Arg Val Arg Asn  
585 590 595

GGT GAA CTC TAT CGC TAC ATT TGT GAC AAA GGA GGT ATT TTT GCG 1844  
Gly Glu Leu Tyr Arg Tyr Ile Cys Asp Lys Gly Gly Ile Phe Ala  
600 605 610

CAG CCT GCA TTT TAT GAA GCA TTT GGG CTT ACA GTT GTT GAA GCC 1889  
Gln Pro Ala Phe Tyr Glu Ala Phe Gly Leu Thr Val Val Glu Ala  
615 620 625

ATG ACC TGT GGT CTT CCC ACA TTT GCT ACC TGC CAC GGT GGT CCA 1934  
Met Thr Cys Gly Leu Pro Thr Phe Ala Thr Cys His Gly Gly Pro  
630 635 640

32

GCT GAG ATT ATA GAA GAC GGT GTT TCA GGA TTT CAT ATC GAT CCA 1979  
Ala Glu Ile Ile Glu Asp Gly Val Ser Gly Phe His Ile Asp Pro  
645 650 655

TAT CAT GCT GAT CAG GCA GAA AAA ATG ACT GAA TTC TTT GTC AAG 2024  
Tyr His Ala Asp Gln Ala Glu Lys Met Thr Glu Phe Phe Val Lys  
660 665 670

TGC AGA GAG GAT CCA AAC TAC TGG ACT AAA ATC TCT GCA GGA GGG 2069  
Cys Arg Glu Asp Pro Asn Tyr Trp Thr Lys Ile Ser Ala Gly Gly  
675 680 685

TTA CTA AGG ATC AAA GAA AGA TAT ACC TGG CAA AAG TAT TCT GAA 2114  
Leu Leu Arg Ile Lys Glu Arg Tyr Thr Trp Gln Lys Tyr Ser Glu  
690 695 700

AGG TTA ATG ACA TTG GCA GGG GTG TAT GGT TTC TGG AAA TAT GTC 2159  
Arg Leu Met Thr Leu Ala Gly Val Tyr Gly Phe Trp Lys Tyr Val  
705 710 715

TCT AAA CTA GAG AGA AGA GAG ACA CGA CGT TAT CTT GAG ATG TTC 2204  
Ser Lys Leu Glu Arg Arg Glu Thr Arg Arg Tyr Leu Glu Met Phe  
720 725 730

TAC ATT TTG AAG TTC CGT GAT CTG GCC AAC TCT GTT CCG CTG GCA 2249  
Tyr Ile Leu Lys Phe Arg Asp Leu Ala Asn Ser Val Pro Leu Ala  
735 740 745

33

ACA GAT GAA GAG CCT TCT ACT ACT GAT GCA GTT GCG ACA TTC CGT 2294  
 Thr Asp Glu Glu Pro Ser Thr Thr Asp Ala Val Ala Thr Phe Arg  
 750 755 760

GGA CCT TGA ACGCTGCTGC TTA CTGAGGT TCCAAGTTGT GTATATATTA. 2343  
 Gly Pro End

CTGTGAAAGG AATAAGTGTA GCTACACAAA AGGTTCTCAA CTATTAGTAT 2393

CTTCTCTGTG TAAATAACGA GAGTGAAAAA TGTAATATTG TTGATGTCTT 2443

GAAACTGAG TTTGCTTTGT TTATTTTAA GTGTATGACA ATATGTATCA 2493

TATAACGGAT TCTTCAGTGA TCATATCAAA AACTACTGAC CATCGAAGTT 2543

AATGAAAATC GACAGCAACA 2563

- 5 These sequences can also be combined together in a suitable plasmid which leads to a combination of the individual characteristics, conditioned by the expression of the protein.
- 10 The promoter should ensure that the foreign gene is expressed in the plant. The promoter can be so chosen that the expression occurs only in specified tissues, at a determined time point in the plant's development or at a time point determined by outside influences. The promoter
- 15 can be homologous or heterologous to the plant. Suitable promoters are e.g. the promoter of the 35S RNA of the cauliflower mosaic virus, the patatin promoter B33 (Rocha-Sosa et al. (1989) EMBO J 8: 23-29) or a promoter

that ensures an expression only in photosynthetically active tissues. Other promoters can be used which ensure an expression only in specified organs, such as the root, tuber, seed, stem or specified cell types such as mesophyllic, epidermal or transport cells.

The coding sequences described herein contain the information for the formation of an mRNA for the large subunit of the ADP-glucose-pyrophosphorylase and the sucrose-phosphate-synthase (SPS) and a part of the information for formation of the small subunit of the ADP-glucose-pyrophosphorylase as well as the sucrose-synthase, that are suitable for the formation of anti-sense RNA to the corresponding genes. Whether a translatable mRNA or an anti-sense nucleic acid is formed, depends on the orientation of the coding sequence in relation to the promoter. If the 3' end of the coding sequence is fused to the 3' end of the promoter, an anti-sense RNA results, and by fusion of the 5' end of the coding to the 3' end of the promoter a translatable RNA results. This latter leads to an increase of the enzyme activity in the cell, whilst the first leads to a reduction of the enzyme activity in the cell.

The coding sequence for the large and small subunit of the ADP-glucose-pyrophosphorylase, the sucrose phosphate synthase and the sucrose synthase can be one of those described in this invention or can be one that is derived by modifications of the sequences described above. Thereby especially modifications of the sequences can be considered which lead to by-passing of the plant's own regulation mechanisms. Modifications to the DNA sequences of the invention can be by known methods, such as e.g. base exchange or targeted or non-targeted mutagenesis. The so-formed derivatives of the DNA sequences of the invention are also within the scope of the invention.

With plasmids, which contain one or more of the DNA sequences of the invention, sugar beet can be transformed with the object of raising and/or reducing the enzyme activity and/or the change of the sucrose concentration.

5

For the introduction of the DNA sequences of the invention in sugar beet, a large number of cloning vectors are available, which contain a replication signal for *E. coli* and a marker, which allows a selection of the transformed cells.

10

According to the introduction method of the desired gene in the plant, other DNA sequences may be suitable. Should the Ti- or Ri-plasmid be used, e.g. for the transformation of the plant cell, then at least the right boundary, often however both the right and left boundary of the Ti- and Ri-Plasmid T-DNA, is attached, as a flanking region, to the gene being introduced. The use of T-DNA for the transformation of plants cells has been intensively researched and is well described in EP 120 516; Hoekama, In: The Binary Plant Vector System, Offset-drukkerij Kanters B.V. Alblaserdam, (1985), Chapter V; Fraley, et al., Crit. Rev. Plant Sci., 4:1-46 and An et al. (1985) EMBO J. 4: 277-287. Once the introduced DNA is integrated in the genome, it is as a rule stable there and remains also in the offspring of the original transformed cells. It normally contains a selection marker, which induces resistance in the transformed plant cells against a biocide or antibiotic such as kanamycin, G 418, bleomycin, hygromycin or phosphinotricin etc. The individual marker employed should therefore allow the selection of transformed cells from cells, which lack the introduced DNA.

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For the introduction of DNA into a plant, besides transformation using *Agrobacteria*, there are many other

techniques available. These techniques include the fusion of protoplasts, microinjection of DNA and electroporation, as well as ballistic methods and virus infection. From the transformed plant material, whole plants can be  
5 regenerated in a suitable medium, which contains antibiotics or biocides for the selection. The resulting plants can then be tested for the presence of introduced DNA. No special demands are placed on the plasmids in injection and electroporation. Simple plasmids, such as  
10 e.g. pUC-derivatives can be used. Should however whole plants be regenerated from such transformed cells the presence of a selectable marker gene is necessary. The transformed cells grow within the plants in the usual manner (see also McCormick et al. (1986) Plant Cell Reports  
15 5: 81-84). These plants can be grown normally and crossed with plants, that possess the same transformed genes or different. The resulting hybrid individuals have the corresponding phenotypical properties.

20

In order to understand the examples forming the basis of this invention all the processes necessary for these tests and which are known per se will first of all be listed:

25 1. Cloning process

The vectors pUC 18/19 and M13mp10 series (Yanisch-Perron et al. (1985) Gene 33: 103-119), as well as the vector EMBL 3 (Frischauf et al. (1983) J Mol Biol 170: 827- 842) were used for cloning.

30

For the plant transformations, the gene constructs were cloned in the binary vector BIN 19 (Bevan (1984) Nucl. Acids Res 12: 8711-8720)

35

2. Bacterial strains

The *E. coli* strain BMH71-18 (Messing et al., Proc. Natl. Acad. Sci. USA (1977), 24, 6342-6346) or TB1 was used for the pUC and M13 mP vectors.

- 5 For the vector BIN19, the *E. coli* strain TB1 exclusively, was used. TB1 is a recombinant-negative, tetracycline-resistant derivative of strain JM101 (Yanisch-Perron et al., Gene (1985), 33, 103-119). The genotype of the TB1 strain is (Bart Barrel, personal communication):
- 10 F'(traD36, proAB, lacI, lacZAM15),  $\Delta$ (lac, pro), SupE, thiS, recA, Sr1::Tn10(TcR).

- The transformation of the plasmids into the potato plants was carried out using *Agrobacterium tumefaciens* strain
- 15 LBA4404 (Bevan, (1984), Nucl. Acids Res. 12, 8711-8720).

### 3. Transformation of *Agrobacterium tumefaciens*

- In the case of BIN19 derivatives, the insertion of the DNA
- 20 into the *Agrobacterium* was effected by direct transformation in accordance with the method of Holsters et al., (1978) (Mol Gene Genet 163: 181-187). The plasmid DNA of the transformed *Agrobacterium* was isolated in accordance with the method of Birnboim and Doly (1979)
- 25 (Nucl Acids Res 7: 1513-1523) and was analysed by gel electrophoresis after suitable restriction cleavage.

### 4. Sucrose phosphate-synthase activity test

- 30 The sucrose phosphate-synthase activity was determined according to the method of Siegel and Stitt (1990, Plant Science 66: 205-210) in a two stage analysis. To 180  $\mu$ l of a solution of 50mM HEPES/KOH (pH 7.4), 5mM magnesium chloride, 5mM fructose-6-phosphate, 25mM
- 35 glucose-6-phosphate and 6mM uridine-5'-diphosphoglucose, 20  $\mu$ l of probe was added and incubated for 10 minutes at

25°C. It was heated for 3 minutes at 95°C, to complete the reaction. After centrifuging, the supernatant was spectroscopically analysed for the liberation of uridine-5'-diphosphate, whereby a pyruvate-kinase coupling enzyme reaction was used. Preparations without hexose phosphate, as well as the measurement of the recovery of added uridine-5'-diphosphate act as controls.

### Examples

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#### Example 1

Cloning of cDNA to large and small subunits of the ADP glucose pyrophosphorylase of sugar beet.

15

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20). Resulting from poly-A+-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were plated out for infection of a bacterial colony and then each is probed with the total cDNA fragment of the large and/or subunit of the AGPase of potato (Müller-Roeber et

30  
35



al., 1990, MGG 224, 136-146) as an EcoRI-fragment. The recombinant phages corresponding to the hybridising signal were isolated. By *in vivo* excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded  
5 cDNA as insertion. The plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

10

#### Example 2

15 Cloning of cDNA to sucrose-phosphate-synthase (SPS) from sugar beet.

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20).  
20 Resulting from poly-A+-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first  
25 cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal  
30 XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were  
35 plated out for infection of a bacterial colony and then each is probed with the total cDNA fragment of the

sucrose-phosphate-synthase (SPS) from spinach (Sonnewald, 1992, Planta) as NotI. The recombinant phages corresponding to the hybridising signal were isolated. By in vivo excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded cDNA as insertion. The plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

### Example 3

Cloning of cDNA to sucrose-synthase from sugar beet.

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20). Resulting from poly-A+-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were plated out for infection of a bacterial colony and then parallel is probed with both EcoRI/BglIII sub fragments sucrose synthase from maize (Worrell et al., 1991, Plant

Cell 3, 1121-1130). The recombinant phages corresponding to the hybridising signal were isolated. By *in vivo* excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded cDNA as insertion. The  
5 plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

10 Example 4

Determination of the nucleotide sequence of the ADP  
glucose pyrophosphorylase, the sucrose synthase and the  
sucrose phosphate synthase of sugar beet and derivation of  
15 the corresponding amino acid sequences

The nucleotide sequences of the insertions obtained from Examples 1 -3, were determined by standard methods by means of the dideoxy method (Sanger et al. (1977) Proc.  
20 Natl. Acad. Sci. USA, 74, 5463-5467). The nucleotide sequences and the amino acid sequences derived therefrom are given in the sequence protocols Seq. ID No. 1-4.

The sequences are shown earlier; the protocols are as  
25 follows:

42

SEQ ID NO: 1

SEQUENCE TYPE: Nucleotide with corresponding protein.

SEQUENCE LENGTH: 1924 base pairs

5 STRANDEDNESS: single

TOPOLOGY: linear

MOLECULE TYPE: cDNA

## ORIGINAL SOURCE

10 ORGANISM: *Beta vulgaris*IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda  
zap

## FEATURES:

15 from 206 to 1770 coding region

PROPERTIES: ADP-glucose-pyrophosphorylase, large subunit

20 SEQ ID NO: 2

SEQUENCE TYPE: Nucleotide with corresponding protein

SEQUENCE LENGTH: 1763 base pairs

STRANDEDNESS: single

25 TOPOLOGY: linear

MOLECULE TYPE: cDNA

## ORIGINAL SOURCE

ORGANISM: *Beta vulgaris*30 IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda  
zap

## FEATURES:

from 3 to 1469 coding region

35

PROPERTIES: ADP-glucose-pyrophosphorylase, small subunit

43

SEQ ID NO: 3

SEQUENCE TYPE: Nucleotide with corresponding protein .

SEQUENCE LENGTH: 3635 base pairs

5 STRANDEDNESS: single

TOPOLOGY: linear

MOLECULE TYPE: cDNA

## ORIGINAL SOURCE

10 ORGANISM: Beta vulgarisIMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda  
zap

## FEATURES:

15 from 31 to 3164 coding region

PROPERTIES: Sucrose-phosphate-synthase

20 SEQ ID NO: 4

SEQUENCE TYPE: Nucleotide with corresponding protein

SEQUENCE LENGTH: 2563 base pairs

STRANDEDNESS: single

25 TOPOLOGY: linear

MOLECULE TYPE: cDNA

## ORIGINAL SOURCE

ORGANISM: Beta vulgaris30 IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda  
zap

## FEATURES:

from 3 to 2300 coding region

35

PROPERTIES: Sucrose synthase

CLAIMS

1. DNA sequence with the coding region for the large subunit of ADP-glucose-pyrophosphorylase for the preparation of sugar beet with changed sucrose concentration, characterised in that the sequence has the following nucleotide sequence (Seq. ID No.1):

CAAAAGAAAA	ACTTCCCATT	TCTACTTCTT	TGCACAATAT	AATTTCCCAC	0050
CAATTTTCT	TTAAATTTCT	CACTTTCATT	TAATCAGTTT	TCAGCAACAT	0100
TCTGATACTC	GACAACCCAC	TTTCTGTTCT	CCCAAGATTC	CAAACCTCTG	0150
ATTCTCATTC	CACTAATATT	TTTGCTTATT	TTTTTCTGG	ATTTAAAGAA	0200
AAGCT ATG GAT GCA AGT GCA GCA GCC ATA AAT GTC AAT GCC CAT	0243				
Met Asp Ala Ser Ala Ala Ala Ile Asn Val Asn Ala His					
5	10				
TTA ACA GAA GTT GGA AAG AAA CGT TTT TTA GGA GAG AGA ATC AGT	0288				
Leu Thr Glu Val Gly Lys Lys Arg Phe Leu Gly Glu Arg Ile Ser					
15	20	25			
CAA AGT TTG AAG GGT AAA GAT CTG AGA GCT CTG TTT TCA AGA ACT	0333				
Gln Ser Leu Lys Gly Lys Asp Leu Arg Ala Leu Phe Ser Arg Thr					
30	35	40			
GAG AGC AAG GGT AGA AAT GTC AAT AAA CCT GGG GTT GCA TTT TCT	0378				
Glu Ser Lys Gly Arg Asn Val Asn Lys Pro Gly Val Ala Phe Ser					
45	50	55			
GTT CTC ACC TCA GAT TTT AAT CAA AGT GTT AAA GAA TCT TTG AAA	0423				
Val Leu Thr Ser Asp Phe Asn Gln Ser Val Lys Glu Ser Leu Lys					
60	65	70			

45

TAT GAG CCA GCA TTA TTT GAA TCT CCA AAA GCT GAC CCA AAA AAT 0468

Tyr Glu Pro Ala Leu Phe Glu Ser Pro Lys Ala Asp Pro Lys Asn

75

80

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GTG GCT GCA ATT GTG CTG GGT GGT GGT GCT GGG ACT CGC CTC TTT 0513

Val Ala Ala Ile Val Leu Gly Gly Gly Ala Gly Thr Arg Leu Phe

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100

CCT CTT ACT AGC AGG AGA GCT AAG CCA GCA GTG CCA ATT GGA GGG 0558

Pro Leu Thr Ser Arg Arg Ala Lys Pro Ala Val Pro Ile Gly Gly

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115

TGT TAC AGG CTG ATT GAT GTG CCT ATG AGC AAC TGC ATC AAC AGT 0603

Cys Tyr Arg Leu Ile Asp Val Pro Met Ser Asn Cys Ile Asn Ser

120

125

130

GGC ATT AGA AAG ATT TTC ATT CTT ACC CAG TTC AAT TCG TTT TCG 0648

Gly Ile Arg Lys Ile Phe Ile Leu Thr Gln Phe Asn Ser Phe Ser

135

140

145

CTT AAT CGT CAT CTT GCT CGA ACC TAT AAT TTT GGA GAT GGT GTG 0693

Leu Asn Arg His Leu Ala Arg Thr Tyr Asn Phe Gly Asp Gly Val

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155

160

AAT TTT GGG GAT GGC TTT GTG GAG GTT TTT GCT GCT ACA CAA ACA 0738

Asn Phe Gly Asp Gly Phe Val Glu Val Phe Ala Ala Thr Gln Thr

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175

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CCT GGA GAA TCA GGA AAG AAA TGG TTC CAG GGC ACC GCT GAT GCA 0783  
Pro Gly Glu Ser Gly Lys Lys Trp Phe Gln Gly Thr Ala Asp Ala  
180 185 190

GTA AGA CAG TTT TTC TGG GCA TTT GAG GAT TCC AAA TCC AAG GAT 0828  
Val Arg Gln Phe Phe Trp Ala Phe Glu Asp Ser Lys Ser Lys Asp  
195 200 205

GTC GAG CAT ATA GTT ATT TTA TCC GGT GAT CAT CTT TAC CGA ATG 0873  
Val Glu His Ile Val Ile Leu Ser Gly Asp His Leu Tyr Arg Met  
210 215 220

GAT TAC ATG AGT TTT TGG CAG AAG CAC ATT GAC ACC AAT GCT GAT 0918  
Asp Tyr Met Ser Phe Trp Gln Lys His Ile Asp Thr Asn Ala Asp  
225 230 235

ATT ACA GTG TCA TGC ATA CCC ATG GAT GAC AGC CGT GCA TCG GAT 0963  
Ile Thr Val Ser Cys Ile Pro Met Asp Asp Ser Arg Ala Ser Asp  
240 245 250

TAT GGG CTG ATG AAG ATT GAT CAC ACT GGA CGC ATT GTC CAT TTT 1008  
Tyr Gly Leu Met Lys Ile Asp His Thr Gly Arg Ile Val His Phe  
255 260 265

GCA GAA AAA CCC AAG GGT TCT GAT CTA ACA GCA ATG CAA GTA GAT 1053  
Ala Glu Lys Pro Lys Gly Ser Asp Leu Thr Ala Met Gln Val Asp  
270 275 280



47

ACA ACT GTT CTT GGG CTC TCT GAC CTT GAA GCT ATG TCA AAT CCA 1098  
Thr Thr Val Leu Gly Leu Ser Asp Leu Glu Ala Met Ser Asn Pro

285

290

295

TAT ATT GCA TCA ATG GGT GTT TAT GTC TTT CGA ACG GAT GTT CTT 1143  
Tyr Ile Ala Ser Met Gly Val Tyr Val Phe Arg Thr Asp Val Leu

300

305

310

ATG GAG CTT CTC AAT CGA AAA TAC CCT TCA AGC AAT GAT TTT GGC 1188  
Met Glu Leu Leu Asn Arg Lys Tyr Pro Ser Ser Asn Asp Phe Gly

315

320

325

TCT GAG ATT ATT CCT TCA GCT GTA GGA GAG TCT AAT GTT CAG GCA 1233  
Ser Glu Ile Ile Pro Ser Ala Val Gly Glu Ser Asn Val Gln Ala

330

335

340

TAT CTA TTT AAT GAC TAC TGG GAG GAT ATC GGA ACC ATA AAG TCT 1278  
Tyr Leu Phe Asn Asp Tyr Trp Glu Asp Ile Gly Thr Ile Lys Ser

345

350

355

TTC TTT GAT TCC AAT TTG GCC CTT ACA CAA CAG CCT CCC AAG TTT 1323  
Phe Phe Asp Ser Asn Leu Ala Leu Thr Gln Gln Pro Pro Lys Phe

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GAA TTC TAC GAT CCA AAA ACA CCT TTT TAT ACA TCT GCA AGA TTT 1368  
Glu Phe Tyr Asp Pro Lys Thr Pro Phe Tyr Thr Ser Ala Arg Phe

375

380

385

48

CTG CCT CCT ACA AAA GTC GAC AGG TGC AAG ATT GTC GAT TCC ATT 1413  
Leu Pro Pro Thr Lys Val Asp Arg Cys Lys Ile Val Asp Ser Ile  
390 395 400

GTA TCC CAT GGT TGT TTT CTA CAG GAG TCT AGC ATC CAA CAT TCC 1458  
Val Ser His Gly Cys Phe Leu Gln Glu Ser Ser Ile Gln His Ser  
405 410 415

ATT GTT GGT GTT CGC TCA AGA TTA GAG TCC GGG GTT GAG TTC CAG 1503  
Ile Val Gly Val Arg Ser Arg Leu Glu Ser Gly Val Glu Phe Gln  
420 425 430

GAC ACC ATG ATG ATG GGC GCA GAT TAC TAT CAA ACT GAA TCA GAA 1548  
Asp Thr Met Met Met Gly Ala Asp Tyr Tyr Gln Thr Glu Ser Glu  
435 440 445

ATT GCT TCT CTG CTT GCT GAG GGA AAG GTT CCT GTT GGT GTC GGA 1593  
Ile Ala Ser Leu Leu Ala Glu Gly Lys Val Pro Val Gly Val Gly  
450 455 460

CAG AAT ACC AAA ATA AAG AAT TGC ATA ATT GAC AAG AAC GCC AAA 1638  
Gln Asn Thr Lys Ile Lys Asn Cys Ile Ile Asp Lys Asn Ala Lys  
465 470 475

ATT GGA AAA GAT GTG GTA ATC GCA AAC ACG GAT GGT GTT GAG GAA 1683  
Ile Gly Lys Asp Val Val Ile Ala Asn Thr Asp Gly Val Glu Glu  
480 485 490

49

GCA GAT AGA CCA AAT GAA GGC TTT TAC ATC AGG TCG GGC ATT ACC 1728  
 Ala Asp Arg Pro Asn Glu Gly Phe Tyr Ile Arg Ser Gly Ile Thr  
 495 500 505

ATC ATT TTG AAG AAC GCA ACC ATA CAA GAC GGT CTT GTG ATT TAG 1773  
 Ile Ile Leu Lys Asn Ala Thr Ile Gln Asp Gly Leu Val Ile End  
 510 515 520

ATTTAATCAT AACCTCATTA GAAAGAAATA ATTTTGCATG ATTTCTTTTT 1823

CATGTAACCT AAAGTGGCTA AACCACGAGG TTTTCTCATC TGTATATATA 1873

ATATGTCTAT AACTATGGAT AATCTTAATA AAAAAAAAAA AAAAAAAAAA 1923

A 1924

2. DNA sequence with the coding region for the small subunit of the ADP-glucose-pyrophosphatase for the preparation of sugar beet with changed sucrose concentration, characterised in that the sequence has the following nucleotide sequence (Seq. ID No.2):

GG ATA ACT GTG CCA TCA ACC TCC TCA AAG AAC CTC CAA AAT AGC 0044  
 Ile Thr Val Pro Ser Thr Ser Ser Lys Asn Leu Gln Asn Ser  
 5 10

CTC GCA TTC TCC TCT TCT TCT CTC TCC GGC GAC AAA ATT CAA ACG 0089  
 Leu Ala Phe Ser Ser Ser Ser Leu Ser Gly Asp Lys Ile Gln Thr  
 15 20 25

ACG TCA TTT CTC AAC CGC CGA TAT TGT AGA ATC TCT TCT AGA GCT 0134  
 Thr Ser Phe Leu Asn Arg Arg Tyr Cys Arg Ile Ser Ser Arg Ala  
 30 35 40

50

CCG ATT GTT GTC TCT CCC AAA GCT GTT TCT GAT TCT AAG AAT TCG 0179  
Pro Ile Val Val Ser Pro Lys Ala Val Ser Asp Ser Lys Asn Ser  
45 50 55

CAG ACT TGT CTT GAC CCT GAA GCC AGC CGT AGT GTT CTT GGT ATT 0224  
Gln Thr Cys Leu Asp Pro Glu Ala Ser Arg Ser Val Leu Gly Ile  
60 65 70

ATA CTT GGA GGT GGT GCT GGT ACA CGT CTT TAC CCG TTG ACT AAG 0269  
Ile Leu Gly Gly Gly Ala Gly Thr Arg Leu Tyr Pro Leu Thr Lys  
75 80 85

AAG AGA GCC AAA CCA GCC GTG CCA CTC GGT GCT AAT TAT AGG CTT 0314  
Lys Arg Ala Lys Pro Ala Val Pro Leu Gly Ala Asn Tyr Arg Leu  
90 95 100

ATT GAT ATC CCA GTG AGC AAT TGT TTG AAC AGT AAT ATT TCC AAA 0359  
Ile Asp Ile Pro Val Ser Asn Cys Leu Asn Ser Asn Ile Ser Lys  
105 110 110

ATA TAT GTT CTT ACA CAA TTC AAT TCT GCT TCT CTG AAT CGT CAT 0404  
Ile Tyr Val Leu Thr Gln Phe Asn Ser Ala Ser Leu Asn Arg His  
115 120 125

CTT TCG CGG GCA TAT GCT AGC AAC ATG GGA GGA TAC AAA AAT GAG 0449  
Leu Ser Arg Ala Tyr Ala Ser Asn Met Gly Gly Tyr Lys Asn Glu  
130 135 140

GGG TTT GTA GAA GTT CTT GCT GCT CAG CAA AGT CCA GAG AAT CCA 0494  
Gly Phe Val Glu Val Leu Ala Ala Gln Gln Ser Pro Glu Asn Pro  
145 150 155

51

AAC TGG TTT CAG GGT ACA GCT GAT GCT GTT AGG CAA TAT CTG TGG 0539  
Asn Trp Phe Gln Gly Thr Ala Asp Ala Val Arg Gln Tyr Leu Trp  
160 165 170

CTT TTC GAA GAG CAC AAT GTT CTT GAG TAC TTG ATT CTT GCT GGT 0584  
Leu Phe Glu Glu His Asn Val Leu Glu Tyr Leu Ile Leu Ala Gly  
175 180 185

GAC CAT TTG TAT CGA ATG GAT TAT GAA AGA TTT GTC CAA GCT CAC 0629  
Asp His Leu Tyr Arg Met Asp Tyr Glu Arg Phe Val Gln Ala His  
190 195 200

AGA GAA ACT GAT GCA GAC ATT ACT GTT GCT GCA TTG CCA ATG GAT 0674  
Arg Glu Thr Asp Ala Asp Ile Thr Val Ala Ala Leu Pro Met Asp  
205 210 215

GAA AAG CGT GCT ACT GCA TTT GGT TTG ATG AAA ATT GAT GAA GAA 0719  
Glu Lys Arg Ala Thr Ala Phe Gly Leu Met Lys Ile Asp Glu Glu  
220 225 230

GGA AGA ATT ATT GAG TTT GCC GAG AAA CCG AAA GGA GAA CAA TTG 0764  
Gly Arg Ile Ile Glu Phe Ala Glu Lys Pro Lys Gly Glu Gln Leu  
235 240 245

AAA GCT ATG AAG GTT GAT ACC ACA ATC CTG GGT CTG GAC GAT GAG 0809  
Lys Ala Met Lys Val Asp Thr Thr Ile Leu Gly Leu Asp Asp Glu  
250 255 260

52

AGA GCA AAA GAA ATG CCA TTC ATA GCC AGC ATG GGC ATA TAT GTT 0854  
Arg Ala Lys Glu Met Pro Phe Ile Ala Ser Met Gly Ile Tyr Val  
265 270 275

ATT AGC AAA GAT GTA ATG CTT AAT CTG CTT CGG GAG CAA TTT CCT 0899  
Ile Ser Lys Asp Val Met Leu Asn Leu Leu Arg Glu Gln Phe Pro  
280 285 290

GGT GCT AAT GAT TTT GGA AGT GAA GTT ATT CCA GGC GCC ACT TCC 0944  
Gly Ala Asn Asp Phe Gly Ser Glu Val Ile Pro Gly Ala Thr Ser  
295 300 305

ATA GGG TTG AGA GTC CAA GCT TAT TTG TAT GAT GGT TAC TGG GAG 0989  
Ile Gly Leu Arg Val Gln Ala Tyr Leu Tyr Asp Gly Tyr Trp Glu  
310 315 320

GAT ATT GGT ACC ATT GAA GCT TTT TAC AAT GCT AAC TTG GGA ATC 1034  
Asp Ile Gly Thr Ile Glu Ala Phe Tyr Asn Ala Asn Leu Gly Ile  
325 330 335

ACC AAA AAG CCG GTG CCA GAT TTT AGC TTC TAT GAT CGT TCA TCT 1079  
Thr Lys Lys Pro Val Pro Asp Phe Ser Phe Tyr Asp Arg Ser Ser  
340 345 350

CCA ATT TAT ACA CAA CCT CGG TAT TTG CCT CCT TCA AAG ATG CTT 1124  
Pro Ile Tyr Thr Gln Pro Arg Tyr Leu Pro Pro Ser Lys Met Leu  
355 360 365

53

GAT GCT GAT ATA ACT GAC AGC GTC ATC GGT GAA GGC TGT GTT ATT 1169  
Asp Ala Asp Ile Thr Asp Ser Val Ile Gly Glu Gly Cys Val Ile  
370 375 380

AAG AAC TGT AAG ATT CAT CAT TCT GTT ATC GGA CTT CGA TCT TGT 1214  
Lys Asn Cys Lys Ile His His Ser Val Ile Gly Leu Arg Ser Cys  
385 390 395

ATC TCG GAG GGT GCA ATC ATT GAG GAC ACA CTG TTG ATG GGA GCT 1259  
Ile Ser Glu Gly Ala Ile Ile Glu Asp Thr Leu Leu Met Gly Ala  
400 405 410

GAT TAT TAT GAG ACT GAT GCT GAT CGG AAA TTC CTG GCT GCT AAG 1304  
Asp Tyr Tyr Glu Thr Asp Ala Asp Arg Lys Phe Leu Ala Ala Lys  
415 420 425

GGT AGT GTA CCT ATT GGA ATT GGG AAT GCA CGT ATT GGG GAT GAT 1349  
Gly Ser Val Pro Ile Gly Ile Gly Asn Ala Arg Ile Gly Asp Asp  
430 435 440

GTC AAG ATT ATC AAC AGT GAC AAT GTA CAA GAA GCA GCA AGA GAA 1394  
Val Lys Ile Ile Asn Ser Asp Asn Val Gln Glu Ala Ala Arg Glu  
445 450 455

ACA GAC GGA TAC TTC ATA AAG AGC GGA ATA GTC ACT ATA ATC AAG 1439  
Thr Asp Gly Tyr Phe Ile Lys Ser Gly Ile Val Thr Ile Ile Lys  
460 465 470

54

GAC GCC ATG ATT CCA AGT GGA ACT GTA ATC TAG AAATGGAGCA 1482  
 Asp Ala Met Ile Pro Ser Gly Thr Val Ile End  
 475 480 485

TATAATAAAT ATCACTGCCT ATTTACAGTA CCTATCTGAG TCTCCCACCA 1532

TGACCCTTTG ATTCAATCTT TTAGTTATGT AAATATTTTT GGCTTTTGCG 1582

ATTTTGCCAT AAATTTGAAG AAGCGAGGAT TCAGGGACGA TAGTGCTATG 1632

AATTGGAAGA AAGGATTG GGGATATCTT TGTAAGACA TTTGACTAC 1682

TGGGCACTAA AAATTTGGTA ATGCTATACC AAAATATATA AAAAGATCTT 1732

GCTGGGTTTT GGTAACAAAAA AAAAAAAAAA A 1763

3. DNA sequence with the coding region for sucrose  
 phosphate - synthase for the preparation of sugar  
 beet with changed sucrose concentration,  
 characterised in that the sequence has the following  
 5 nucleotide sequence (Seq. ID No.3):

GGGCTGCAGG GAAGCTCTGA ACTTCAAAA ATG GCG GGA AAT GAT 0044  
 Met Ala Gly Asn Asp  
 5

TGG ATA AAC AGT TAT TTA GAG GCA ATT CTG GAT GTG GGT CCA GGA 0089  
 Trp Ile Asn Ser Tyr Leu Glu Ala Ile Leu Asp Val Gly Pro Gly  
 10 15 20

CTT GAT GAT GCA AAA TCA TCT TTG CTT TTG AGA GAA AGA GGC AGG 0134  
 Leu Asp Asp Ala Lys Ser Ser Leu Leu Leu Arg Glu Arg Gly Arg  
 25 30 35



55

TTT AGT CCT ACT CGT TAC TTT GTT GAA GAA GTT ATC ACT GGT TTT 0179  
 Phe Ser Pro Thr Arg Tyr Phe Val Glu Glu Val Ile Thr Gly Phe

40

45

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GAT GAA ACC GAC CTT CAT CGT TCA TGG GTT CGG GCA CAA GCA ACA 0224  
 Asp Glu Thr Asp Leu His Arg Ser Trp Val Arg Ala Gln Ala Thr

55

60

65

AGG AGT CCT CAA GAG AGG AAT ACT AGA TTG GAG AAC ATG TGT TGG 0269  
 Arg Ser Pro Gln Glu Arg Asn Thr Arg Leu Glu Asn Met Cys Trp

70

75

80

AGA ATT TGG AAT TTG GCT CGT CAG AAG AAG CAG CTT GAG AAT GAA 0314  
 Arg Ile Trp Asn Leu Ala Arg Gln Lys Lys Gln Leu Glu Asn Glu

85

90

95

GAA GCT CAG CGG AAG ACA AAA CGT CGT ATG GAG CTT GAG AGG GGT 0359  
 Glu Ala Gln Arg Lys Thr Lys Arg Arg Met Glu Leu Glu Arg Gly

100

105

110

CGT CGA GAA GCA ACT GCT GAT ATG TCG GAG GAC TTA TCA GAA GGC 0404  
 Arg Arg Glu Ala Thr Ala Asp Met Ser Glu Asp Leu Ser Glu Gly

115

120

125

GAA AAG GAC ATT TCA GCT CAT GGT GAT AGC ACC CGT CCT AGA TTG 0449  
 Glu Lys Asp Ile Ser Ala His Gly Asp Ser Thr Arg Pro Arg Leu

130

135

140

CCA AGA ATA AAT TCT CTT GAT GCT ATG GAG ACA TGG ATT AGT CAA 0494  
 Pro Arg Ile Asn Ser Leu Asp Ala Met Glu Thr Trp Ile Ser Gln

145

150

155

56

CAA AAG GAA AAA AAA CTC TAC CTT GTT TTG ATA AGT CTT CAT GGT 0539  
Gln Lys Glu Lys Lys Leu Tyr Leu Val Leu Ile Ser Leu His Gly  
160 165 170

TTG ATA CGA GGT GAA AAC ATG GAA CTT GGC CGT GAT TCT GAT ACT 0584  
Leu Ile Arg Gly Glu Asn Met Glu Leu Gly Arg Asp Ser Asp Thr  
175 180 185

GGT GGT CAG GTT AAG TAT GTG GTT GAG CTT GCA AGG GCT CTA GGT 0629  
Gly Gly Gln Val Lys Tyr Val Val Glu Leu Ala Arg Ala Leu Gly  
190 195 200

TCG ATG CCA GGT GTT TAT AGA GTT GAT TTG CTA ACT AGG CAA GTT 0674  
Ser Met Pro Gly Val Tyr Arg Val Asp Leu Leu Thr Arg Gln Val  
205 210 215

TCA TCT CCT GAC GTG GAT TGG AGT TAT GGG GAG CCT ACT GAG ATG 0719  
Ser Ser Pro Asp Val Asp Trp Ser Tyr Gly Glu Pro Thr Glu Met  
220 225 230

CTG AAT CCA AGG GAT TCC AAT GGT TTT GAT GAT GAT GAT GAT GAA 0764  
Leu Asn Pro Arg Asp Ser Asn Gly Phe Asp Asp Asp Asp Asp Glu  
235 240 245

ATG GGA GAG AGT AGT GGT GCT TAC ATT GTT CGT ATA CCA TTT GGG 0809  
Met Gly Glu Ser Ser Gly Ala Tyr Ile Val Arg Ile Pro Phe Gly  
250 255 260

57

CCG AGG GAT AAG TAT ATC GCA AAA GAA GAG CTT TGG CCC TAT ATT 0854  
Pro Arg Asp Lys Tyr Ile Ala Lys Glu Glu Leu Trp Pro Tyr Ile  
265 270 275

CCT GAA TTT GTT GAT GGT GCT CTA AAC CAC ATA GTT CAA ATG TCC 0899  
Pro Glu Phe Val Asp Gly Ala Leu Asn His Ile Val Gln Met Ser  
280 285 290

AAA GTT TTA GGT GAG CAA ATT GGT AGC GGG GAA ACA GTT TGG CCA 0944  
Lys Val Leu Gly Glu Gln Ile Gly Ser Gly Glu Thr Val Trp Pro  
295 300 305

GTT GCC ATT CAT GGA CAT TAT GCT GAT GCT GGT GAT TCT GCT GCT 0989  
Val Ala Ile His Gly His Tyr Ala Asp Ala Gly Asp Ser Ala Ala  
310 315 320

CTT CTT TCT GGT GGC CTA AAT GTT CCA ATG CTT TTA ACG GGG CAT 1034  
Leu Leu Ser Gly Gly Leu Asn Val Pro Met Leu Leu Thr Gly His  
325 330 335

TCT CTT GGC CGA GAC AAG TTA GAG CAG CTC CTC AAA CAG GGT CGA 1079  
Ser Leu Gly Arg Asp Lys Leu Glu Gln Leu Leu Lys Gln Gly Arg  
340 345 350

ATG TCT AAA GAT GAC ATA AAC AAT ACA TAC AAA ATA ATG CGT AGG 1124  
Met Ser Lys Asp Asp Ile Asn Asn Thr Tyr Lys Ile Met Arg Arg  
355 360 365

58

ATA GAA GCC GAA GAG TTA TCA CTT GAT GCC TCT GAG ATA GTC ATA 1169  
Ile Glu Ala Glu Glu Leu Ser Leu Asp Ala Ser Glu Ile Val Ile  
370 375 380

ACT AGT ACA AGA CAA GAA ATA GAA GAG CAA TGG CAC CTC TAT GAT 1214  
Thr Ser Thr Arg Gln Glu Ile Glu Glu Gln Trp His Leu Tyr Asp  
385 390 395

GGG TTT GAT CCT GTG CTA GAA CGT AAA CTC CGT GCT AGG ATG AAG 1259  
Gly Phe Asp Pro Val Leu Glu Arg Lys Leu Arg Ala Arg Met Lys  
400 405 410

CGT GGT GTA AGC TGT TAT GGA AGG TTC ATG CCC CGG ATG GTT GTT 1304  
Arg Gly Val Ser Cys Tyr Gly Arg Phe Met Pro Arg Met Val Val  
415 420 425

ATT CCT CCT GGA ATG GAA TTC AAT CAT ATT GTT CCA CAT GAG GGT 1349  
Ile Pro Pro Gly Met Glu Phe Asn His Ile Val Pro His Glu Gly  
430 435 440

GAT ATG GAT GGT GAA ACA GAA GAA ACT GAA GAG CAT CCT ACA TCA 1394  
Asp Met Asp Gly Glu Thr Glu Glu Thr Glu Glu His Pro Thr Ser  
445 450 455

CCT GAT CCA CCT ATC TGG GCT GAG ATT ATG CGC TTC TTT TCT AAA 1439  
Pro Asp Pro Pro Ile Trp Ala Glu Ile Met Arg Phe Phe Ser Lys  
460 465 470

59

CCA AGG AAG CCA ATG ATA CTT GCC CTT GCT AGG CCT GAC CCG AAG 1484  
Pro Arg Lys Pro Met Ile Leu Ala Leu Ala Arg Pro Asp Pro Lys  
475 480 485

AAG AAT ATC ACG ACT TTG GTC AAA GCA TTT GGA GAA TGC CGT CCA 1529  
Lys Asn Ile Thr Thr Leu Val Lys Ala Phe Gly Glu Cys Arg Pro  
490 495 500

CTA AGG GAG CTA GCT AAT CTT ACT CTT ATA ATG GGT AAC CGA GAT 1574  
Leu Arg Glu Leu Ala Asn Leu Thr Leu Ile Met Gly Asn Arg Asp  
505 510 515

GGT ATT GAC GAG ATG TCA AGC ACC AGT TCT TCA GTT CTC CTG TCA 1619  
Gly Ile Asp Glu Met Ser Ser Thr Ser Ser Ser Val Leu Leu Ser  
520 525 530

GTG CTT AAG CTA ATT GAT CAA TAC GAC CTT TAT GGT CAA GTA GCA 1664  
Val Leu Lys Leu Ile Asp Gln Tyr Asp Leu Tyr Gly Gln Val Ala  
535 540 545

TAC CCC AAA CAT CAC AAG CAA GCT GAT GTT CCT GAG ATT TAT CGT 1709  
Tyr Pro Lys His His Lys Gln Ala Asp Val Pro Glu Ile Tyr Arg  
550 555 600

TTG GCA GCA AAG ACA AAG GGA GTC TTT ATT AAT CCA GCT TTT ATT 1754  
Leu Ala Ala Lys Thr Lys Gly Val Phe Ile Asn Pro Ala Phe Ile  
605 610 615

60

GAG CCA TTT GGG CTG ACT CTA ATA GAG GCA GCA GCT CAT GGT TTA 1799  
Glu Pro Phe Gly Leu Thr Leu Ile Glu Ala Ala Ala His Gly Leu  
620 625 630

CCG ATG GTT GCT ACG AAA AAT GGA GGC CCT GTT GAT ATC CAG AGG 1844  
Pro Met Val Ala Thr Lys Asn Gly Gly Pro Val Asp Ile Gln Arg  
635 640 645

GTC CTT GAT AAT GGT CTT CTT GTG GAT CCT CAT GAG CAG CAG TCT 1889  
Val Leu Asp Asn Gly Leu Leu Val Asp Pro His Glu Gln Gln Ser  
650 655 660

ATT GCT ACT GCT TTG CTG AAG CTT GTT GCT GAT AAG CAA CTA TGG 1934  
Ile Ala Thr Ala Leu Leu Lys Leu Val Ala Asp Lys Gln Leu Trp  
665 670 675

ACA AAA TGC CAG CAA AAT GGA CTG AAA AAT ATT CAT CTC TAC TCT 1979  
Thr Lys Cys Gln Gln Asn Gly Leu Lys Asn Ile His Leu Tyr Ser  
680 685 690

TGG CCA GAG CAT TCG AAG ACA TAC CTA TCT CGA ATA GCC AGT TCG 2024  
Trp Pro Glu His Ser Lys Thr Tyr Leu Ser Arg Ile Ala Ser Ser  
695 700 705

AGA CAA AGG CAA CCA CAG TGG CAA AGA AGT AGT GAT GAA GGG CTT 2069  
Arg Gln Arg Gln Pro Gln Trp Gln Arg Ser Ser Asp Glu Gly Leu  
710 715 720

61

GAC AAT CAA GAG CCT GAA TCT CCA AGT GAT TCT TTA AGA GAT ATA 2114  
Asp Asn Gln Glu Pro Glu Ser Pro Ser Asp Ser Leu Arg Asp Ile  
725 730 735

AAG GAT ATA TCT CTA AAC CTT GAA GTT CTC GTT AGA CCG GAG AAA 2159  
Lys Asp Ile Ser Leu Asn Leu Glu Val Leu Val Arg Pro Glu Lys  
740 745 750

AGG GTG AAG ACG TTG AAA ATC TTG GGA TTG ATG ACA AAA GCA AAT 2204  
Arg Val Lys Thr Leu Lys Ile Leu Gly Leu Met Thr Lys Ala Asn  
755 760 765

TCG AGA ATG CTG TTA TGT TCA TGG TCT AAT GGT GTC CAT AAG ATG 2249  
Ser Arg Met Leu Leu Cys Ser Trp Ser Asn Gly Val His Lys Met  
770 775 780

CTT CGG AAG GCT CGG TTC TCT GAC AAA GTA GAT CAG GCT TCT AGT 2294  
Leu Arg Lys Ala Arg Phe Ser Asp Lys Val Asp Gln Ala Ser Ser  
785 790 795

AAA TAT CCA GCA TTT AGG AGG AGA AAA CTT ATA TAT GTT ATT GCT 2339  
Lys Tyr Pro Ala Phe Arg Arg Arg Lys Leu Ile Tyr Val Ile Ala  
800 805 810

GTA GAC GGG GAT TAT GAA GAT GGA CTT TTT GAT ATT GTT CGG AGG 2384  
Val Asp Gly Asp Tyr Glu Asp Gly Leu Phe Asp Ile Val Arg Arg  
815 820 825

62

ATA TTT GAT GCT GCT GGC AAG GAG AAG ATT GAA GGT TCC ATC GGG 2429  
Ile Phe Asp Ala Ala Gly Lys Glu Lys Ile Glu Gly Ser Ile Gly  
830 835 840

TTT ATA TTG TCA ACA TCC TAT TCT ATG CCC GAA ATT CAG AAC TAT 2474  
Phe Ile Leu Ser Thr Ser Tyr Ser Met Pro Glu Ile Gln Asn Tyr  
845 850 855

TTG CTA TCA AAA GGC TTC AAT CTT CAT GAT TTT GAT GCA TAT ATA 2519  
Leu Leu Ser Lys Gly Phe Asn Leu His Asp Phe Asp Ala Tyr Ile  
860 865 870

TGC AAC AGT GGG AGT GAG TTG TAC TAT TCA TCT TTG AAC TCA GAG 2564  
Cys Asn Ser Gly Ser Glu Leu Tyr Tyr Ser Ser Leu Asn Ser Glu  
875 880 885

GAG AGT AAT ATT ATA GCA GAT TCA GAT TAC CAT TCA CAC ATA GAG 2609  
Glu Ser Asn Ile Ile Ala Asp Ser Asp Tyr His Ser His Ile Glu  
890 895 900

TAC AGA TGG GGT GGA GAA GGC CTT AGA AGG ACT TTG CTT CGC TGG 2654  
Tyr Arg Trp Gly Gly Glu Gly Leu Arg Arg Thr Leu Leu Arg Trp  
905 910 915

GCA GCT TCC ATC ACA GAA AAA AAT GGT GAA AAC GAA GAA CAG GTT 2699  
Ala Ala Ser Ile Thr Glu Lys Asn Gly Glu Asn Glu Glu Gln Val  
920 925 930



63

ATT ACT GAA GAT GAA GAA GTT TCT ACG GGT TAT TGC TTT GCG TTT 2744  
Ile Thr Glu Asp Glu Glu Val Ser Thr Gly Tyr Cys Phe Ala Phe  
935 940 945

AAA ATA AAG AAC CAA AAT AAG GTT CCC CCT ACG AAG GAG CTC CGC 2789  
Lys Ile Lys Asn Gln Asn Lys Val Pro Pro Thr Lys Glu Leu Arg  
950 955 960

AAG TCA ATG AGG ATT CAA GCT CTT CGT TGC CAT GTG ATT TAC TGT 2834  
Lys Ser Met Arg Ile Gln Ala Leu Arg Cys His Val Ile Tyr Cys  
965 970 975

CAG AAC GGA TCT AAA ATG AAT GTG ATT CCA GTA CTA GCA TCC CGT 2879  
Gln Asn Gly Ser Lys Met Asn Val Ile Pro Val Leu Ala Ser Arg  
980 985 990

TCT CAA GCC CTC AGG TAT CTT TAT GTT CGT TGG GGA GTT GAG TTG 2924  
Ser Gln Ala Leu Arg Tyr Leu Tyr Val Arg Trp Gly Val Glu Leu  
995 1000 1005

TCG AAG ATG GTT GTC TTT GTT GGA GAA TGT GGT GAC ACA GAT TAT 2969  
Ser Lys Met Val Val Phe Val Gly Glu Cys Gly Asp Thr Asp Tyr  
1010 1015 1020

GAA GGC TTG CTT GGC GGG GTC CAT AAA ACC GTA ATA CTG AAG GGA 3014  
Glu Gly Leu Leu Gly Gly Val His Lys Thr Val Ile Leu Lys Gly  
1025 1030 1035

64

GTC TCC AAC ACT GCT TTA AGG TCT CTC CAT GCC AAC AGA AGT TAC 3059  
 Val Ser Asn Thr Ala Leu Arg Ser Leu His Ala Asn Arg Ser Tyr  
 1040 1045 1050

CCT CTT TCA CAT GTC GTG TCG CTT GAC AGC CCC AAT ATT GGC GAG 3104  
 Pro Leu Ser His Val Val Ser Leu Asp Ser Pro Asn Ile Gly Glu  
 1055 1060 1065

GTG AGC AAA GGG TGC AGC AGC TCC GAG ATC CAG TCC ATC GTC ACA 3149  
 Val Ser Lys Gly Cys Ser Ser Ser Glu Ile Gln Ser Ile Val Thr  
 1070 1075 1080

AAA CTC TCC AAA GCT TAA TCAGATATCT GCTGCTTTCT TTTGGGTAAG 3197  
 Lys Leu Ser Lys Ala End  
 1085

CAAGGTTTCA TCTTATATGA TTATATCATA AGATACTATA TAAGCACCTT 3247

ATTGGTAAGT CAGTCCCATATAATAATGT ACTTCAGAAC CACAATACTT 3297

AAAAGTTGGT TCAGTAGTGA TTAGTCTCAT AATAATCATA TAATTACACA 3347

TCCGCTGTTA ACTAGTGGTA ATATCTAAGC TCAACAATAA AGATGTAAAA 3397

TGCTAGTATG GAAATGAATT GCTAGCTGTT GATCTCTTTC CCTTTATTCT 3447

GTATTATTTC TTTCCTCATC TCATGTAAAA ACAATTTTCT GAAGGTGTAC 3497

AGTTTTTTCC CCTTATATAT CTGTATTATT TCTACTATTT TTTGTTTGTA 3547

AGAATATCCT CTCATCGAGG AGTGATAATT AAATAACCGG CTGCTAAAT 3597

ATAAAGCTTA TTCGAGTTAA AAAAAAAAAA AAAAAAAAAA

3635

4. DNA sequence with the coding region for  
sucrose-synthase for the preparation of sugar beet  
with modified sucrose concentration, characterised  
in that the sequence has the following nucleotide  
sequence (Seq. ID No.4):

CT GCA GGA GGG AAA CAA ATT CTT AGC GAT GGC CCG TTT AGC GAA 0044  
Ala Gly Gly Lys Gln Ile Leu Ser Asp Gly Pro Phe Ser Glu

5

10

GTT CTT AGG TCT GCT CAG GAA GCA ATA GTT GTT CCT CCC TTT GTT 0089  
Val Leu Arg Ser Ala Gln Glu Ala Ile Val Val Pro Pro Phe Val  
15 20 25

GCT ATA GCA GTC CGT CCA AGA CCT GGA GTT TGG GAA TAT GTT CGT 0134  
Ala Ile Ala Val Arg Pro Arg Pro Gly Val Trp Glu Tyr Val Arg  
30 35 40

GTT AAT GTC TCT GAA CTG AAT GTG GAG CAG CTA ACT GTG TCT GAG 0179  
Val Asn Val Ser Glu Leu Asn Val Glu Gln Leu Thr Val Ser Glu  
45 50 55

TAT CTC CAT TTC AAG GAA GAA CTT GTG GAT GGA AAG GCT GAT GAC 0224  
Tyr Leu His Phe Lys Glu Glu Leu Val Asp Gly Lys Ala Asp Asp  
60 65 70

CAC TAT GTG CTT GAG CTT GAT TTC GAG CCT TTT AAT GAA TCA GTT 0269  
His Tyr Val Leu Glu Leu Asp Phe Glu Pro Phe Asn Glu Ser Val  
75 80 85

CCA CGT CCA ACT CGC TCT TCA TCA ATT GGT AAT GGT GTT CAG TTC 0314  
Pro Arg Pro Thr Arg Ser Ser Ser Ile Gly Asn Gly Val Gln Phe  
90 95 100

66

CTC AAT CGG CAC CTG TCA TCA AGC ATG TTC TGC AAC AAA GAT TGC 0359  
 Leu Asn Arg His Leu Ser Ser Ser Met Phe Cys Asn Lys Asp Cys  
 105 110 115

TTG GAG CCG TTA CTT GAT TTT CTT AGA GTG CAC AAA CAT AAA GGA 0404  
 Leu Glu Pro Leu Leu Asp Phe Leu Arg Val His Lys His Lys Gly  
 120 125 130

GTT GTC ATG ATG TTG AAT GAT CGG ATA CAG ACT ATC CAG CGT CTT 0449  
 Val Val Met Met Leu Asn Asp Arg Ile Gln Thr Ile Gln Arg Leu  
 135 140 145

CAG TCT GCA TTG TCT AAA GCT GAG GAT TAT CTT ATC AAA CTT CCA 0494  
 Gln Ser Ala Leu Ser Lys Ala Glu Asp Tyr Leu Ile Lys Leu Pro  
 150 155 160

GCA GAT ACA CCT TAC TCT GAG TTC GAA TTT GTA ATC CAA GGT ATG 0539  
 Ala Asp Thr Pro Tyr Ser Glu Phe Glu Phe Val Ile Gln Gly Met  
 165 170 175

GGT TTT GAA AGA GGC TGG GGT GAT ACT GCT GAA AGG GTT CTA GAA 0584  
 Gly Phe Glu Arg Gly Trp Gly Asp Thr Ala Glu Arg Val Leu Glu  
 180 185 190

ATG ATG CAT CTA CTA CTA GAT ATC CTT CAG GCT CCC GAT CCG TCT 0629  
 Met Met His Leu Leu Leu Asp Ile Leu Gln Ala Pro Asp Pro Ser  
 195 200 205

67

ACA TTA GAG ACA TTT CTG GGA AGA CTT CCC ATG GTG TTT AAT GTG 0674  
Thr Leu Glu Thr Phe Leu Gly Arg Leu Pro Met Val Phe Asn Val  
210 215 220

GTC ATT TTG TCT GTA CAT GGA TAT TTT GGA CAG GCA CAT GTG CTC 0719  
Val Ile Leu Ser Val His Gly Tyr Phe Gly Gln Ala His Val Leu  
225 230 235

GGC TTG CCT GAC ACT GGT GGG CAG ATA GTT TAT ATA CTT GAC CAA 0764  
Gly Leu Pro Asp Thr Gly Gly Gln Ile Val Tyr Ile Leu Asp Gln  
240 245 250

GTG CGG TCT CTG GAA CAT GAA ATG CTC CAA CGA ATA AAG AAG CAA 0809  
Val Arg Ser Leu Glu His Glu Met Leu Gln Arg Ile Lys Lys Gln  
255 260 265

GGA CTA GAT GTG ACT CCT AGA ATT CTT ATC GTG AGT CGG TTG ATT 0854  
Gly Leu Asp Val Thr Pro Arg Ile Leu Ile Val Ser Arg Leu Ile  
270 275 280

CCT GAC GCT AAA GGG ACC ACG TGC AAT CAA CGT ATG GAG AAA GTC 0899  
Pro Asp Ala Lys Gly Thr Thr Cys Asn Gln Arg Met Glu Lys Val  
285 290 295

AGT GGA ACA GAG CAT GCT AGT ATC CTG AGA GTT CCT TTC CGA TCA 0944  
Ser Gly Thr Glu His Ala Ser Ile Leu Arg Val Pro Phe Arg Ser  
300 305 310

68

GAG AAA GGA ATC CTC CGC AAA TGG ATA TCT AGA TTT GAT GTA TGG 0989  
Glu Lys Gly Ile Leu Arg Lys Trp Ile Ser Arg Phe Asp Val Trp  
315 320 325

CCT TAT TTA GAG ACC TTC ACT GAG GAT GCA GCT GGT GAA ATT ATT 1034  
Pro Tyr Leu Glu Thr Phe Thr Glu Asp Ala Ala Gly Glu Ile Ile  
330 335 340

GGC GAG TTG CAG GGT CGT CCA GAT CTG ATA ATT GGC AAC TAC AGC 1079  
Gly Glu Leu Gln Gly Arg Pro Asp Leu Ile Ile Gly Asn Tyr Ser  
345 350 355

GAT GGG AAT ATA GTT GCT TCT TTA TTG TCC CAC AAA ATG GGT GTC 1124  
Asp Gly Asn Ile Val Ala Ser Leu Leu Ser His Lys Met Gly Val  
360 365 370

ACC CAG TGC AAT ATA GCC CAT GCA TTG GAG AAA ACC AAG TAT CCA 1169  
Thr Gln Cys Asn Ile Ala His Ala Leu Glu Lys Thr Lys Tyr Pro  
375 380 385

GAT TCT GAT ATT TAC TGG AAA AGA TTT GAG GAC AAA TAT CAC TTC 1214  
Asp Ser Asp Ile Tyr Trp Lys Arg Phe Glu Asp Lys Tyr His Phe  
390 395 400

TCG TGT CAA TTT TCA GCT GAC TTG ATG GCA ATG AAT CAT GCT GAT 1259  
Ser Cys Gln Phe Ser Ala Asp Leu Met Ala Met Asn His Ala Asp  
405 410 415

69

TTC ATC ATT ACG AGT ACT TAC CAA GAG ATA GCT GGA ACG AAG AAT 1304  
Phe Ile Ile Thr Ser Thr Tyr Gln Glu Ile Ala Gly Thr Lys Asn  
420 425 430

ACT GTT GGT CAA TAT GAA AGC CAT AAG GCC TTT ACT TTT CCG GGG 1349  
Thr Val Gly Gln Tyr Glu Ser His Lys Ala Phe Thr Phe Pro Gly  
435 440 445

CTG TAT CGG GTG GTT CAC GGG ATT GAT GTC TTT GAT CCC AAG TTT 1394  
Leu Tyr Arg Val Val His Gly Ile Asp Val Phe Asp Pro Lys Phe  
450 455 460

AAT ATT GTC TCG CCA GGG GCA GAC ATG GCC ATC TAC TTC CCA TTT 1439  
Asn Ile Val Ser Pro Gly Ala Asp Met Ala Ile Tyr Phe Pro Phe  
465 470 475

TCA GAG AAG GAT GTC ACC TGT CTC ACT TCA CTT CAT AGA CTT ATA 1484  
Ser Glu Lys Asp Val Thr Cys Leu Thr Ser Leu His Arg Leu Ile  
480 485 490

GAG CAG CTC CTA TTC AAA CCT GAG CAG AAC GAA GAA CAC ATT GGT 1529  
Glu Gln Leu Leu Phe Lys Pro Glu Gln Asn Glu Glu His Ile Gly  
495 500 505

GTA TTA GAT GAT ACC TCA AAG CCA ATT ATA TTT TCC ATG GCG AGG 1574  
Val Leu Asp Asp Thr Ser Lys Pro Ile Ile Phe Ser Met Ala Arg  
510 515 520

70

CTA GAC CGT GTG AAG AAT ATA ACA GGG CTG GTA GAG TGC TAT GGC 1619  
 Leu Asp Arg Val Lys Asn Ile Thr Gly Leu Val Glu Cys Tyr Gly  
 525 530 535

AAG AAT GCG AAA CTC AGG GAA CTG GCA AAC CTG GTT GTA GTG GCT 1664  
 Lys Asn Ala Lys Leu Arg Glu Leu Ala Asn Leu Val Val Val Ala  
 540 545 550

GGG TAC AAT GAT GTA AAA AAG TCG AAT GAC AGG GAG GAA ATT GCC 1709  
 Gly Tyr Asn Asp Val Lys Lys Ser Asn Asp Arg Glu Glu Ile Ala  
 555 560 565

GAA ATC GAG AAG ATG CAC AGG CTT ATA CAG GAG TAT AAT TTA AGA 1754  
 Glu Ile Glu Lys Met His Arg Leu Ile Gln Glu Tyr Asn Leu Arg  
 570 575 580

GGA CAA TTT CGC TGG ATT GCT TCT CAA ACA AAT AGA GTA CGA AAT 1799  
 Gly Gln Phe Arg Trp Ile Ala Ser Gln Thr Asn Arg Val Arg Asn  
 585 590 595

GGT GAA CTC TAT CGC TAC ATT TGT GAC AAA GGA GGT ATT TTT GCG 1844  
 Gly Glu Leu Tyr Arg Tyr Ile Cys Asp Lys Gly Gly Ile Phe Ala  
 600 605 610

CAG CCT GCA TTT TAT GAA GCA TTT GGG CTT ACA GTT GTT GAA GCC 1889  
 Gln Pro Ala Phe Tyr Glu Ala Phe Gly Leu Thr Val Val Glu Ala  
 615 620 625



71

ATG ACC TGT GGT CTT CCC ACA TTT GCT ACC TGC CAC GGT GGT CCA 1934  
Met Thr Cys Gly Leu Pro Thr Phe Ala Thr Cys His Gly Gly Pro  
630 635 640

GCT GAG ATT ATA GAA GAC GGT GTT TCA GGA TTT CAT ATC GAT CCA 1979  
Ala Glu Ile Ile Glu Asp Gly Val Ser Gly Phe His Ile Asp Pro  
645 650 655

TAT CAT GCT GAT CAG GCA GAA AAA ATG ACT GAA TTC TTT GTC AAG 2024  
Tyr His Ala Asp Gln Ala Glu Lys Met Thr Glu Phe Phe Val Lys  
660 665 670

TGC AGA GAG GAT CCA AAC TAC TGG ACT AAA ATC TCT GCA GGA GGG 2069  
Cys Arg Glu Asp Pro Asn Tyr Trp Thr Lys Ile Ser Ala Gly Gly  
675 680 685

TTA CTA AGG ATC AAA GAA AGA TAT ACC TGG CAA AAG TAT TCT GAA 2114  
Leu Leu Arg Ile Lys Glu Arg Tyr Thr Trp Gln Lys Tyr Ser Glu  
690 695 700

AGG TTA ATG ACA TTG GCA GGG GTG TAT GGT TTC TGG AAA TAT GTC 2159  
Arg Leu Met Thr Leu Ala Gly Val Tyr Gly Phe Trp Lys Tyr Val  
705 710 715

TCT AAA CTA GAG AGA AGA GAG ACA CGA CGT TAT CTT GAG ATG TTC 2204  
Ser Lys Leu Glu Arg Arg Glu Thr Arg Arg Tyr Leu Glu Met Phe  
720 725 730

72

TAC ATT TTG AAG TTC CGT GAT CTG GCC AAC TCT GTT CCG CTG GCA 2249  
 Tyr Ile Leu Lys Phe Arg Asp Leu Ala Asn Ser Val Pro Leu Ala  
 735 740 745

ACA GAT GAA GAG CCT TCT ACT ACT GAT GCA GTT GCG ACA TTC CGT 2294  
 Thr Asp Glu Glu Pro Ser Thr Thr Asp Ala Val Ala Thr Phe Arg  
 750 755 760

GGA CCT TGA ACGCTGCTGC TTACTGAGGT TCCAAGTTGT GTATATATTA 2343  
 Gly Pro End

CTGTGAAAGG AATAAGTGTA GCTACACAAA AGGTTCTCAA CTATTAGTAT 2393

CTTCTCTGTG TAAATAACGA GAGTGAAAAA TGTAATATTG TTGATGTCTT 2443

GAAAACTGAG TTGCTTTTGT TTATTTTAA GTGTATGACA ATATGTATCA 2493

TATAACGGAT TCTTCAGTGA TCATATCAAA AACTACTGAC CATCGAAGTT 2543

AATGAAAATC GACAGCAACA 2563

5. Derivatives of DNA sequences according to any one of claims 1 to 4 characterised in that these derivatives are obtained by exchange of single bases or by targeted or non-targeted mutagenesis.

5

6. Plasmids containing

- a) a suitable promoter which ensures that the coding sequence meets a suitable time point or in the specified developments in the transgenic plant or in determined genes of transgenic plants;

10

- 5                   b)    at least one coding sequence according to  
                    claims 1 to 5, that
- i)    is so coupled to the promoter that the  
                          formation of an RNA is allowed which is  
                          into a protein, whereby the protein  
                          demonstrates an enzymatic activity which  
                          leads to a modification of the sucrose  
                          concentration in the plant, or
- 10                   ii)   which is so coupled to the promoter that  
                          the coding part is read, which leads to  
                          the formation of a so-called anti-sense  
                          RNA which under-expresses the formation  
                          of the protein coded from an endogenous  
                          gene in the plant, that is involved in  
15                   the sucrose biosynthesis; and
- c)    A non-coding termination sequence that contains  
                          the signal for the termination and poly-  
                          adenylation of the transcript.
- 20                   7.    Use of the DNA-sequences according to any one of  
                          claims 1 to 6 for the preparation of sugar beet with  
                          changed sucrose concentration.
- 25                   8.    Use of the plasmids according to any one of claims  
                          1-4 for the preparation of derivatives by targeted  
                          or non-targeted mutagenesis
9.    Sugar beet containing a DNA sequence according to  
30                   any one of claims 1-5.

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